# WATER QUALITY AND FECAL COLIFORM DYANAMICS IN AQUACULTURE BHERI OF EAST KOLKATA WETLAND

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**IN**

**MICROBIOLOGY**


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# PU-SI Certificate

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The entire dissertation work has been done under my supervision and this work has not been submitted elsewhere for a degree.

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I hereby declare that the work which is being presented in this dissertation entitled **"Water quality and Faecal coliform dynamics in Aquaculture bheri of East Kolkata Wetland"** in partial fulfilment of the requirements for the award of the degree of Master of Science in Microbiology is an authentic record of my own work carried out during the period from January, 2025 to May, 2025 under the supervision of Dr. Souryadeep Mukherjee, Department of Life Sciences, Presidency University.

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Date: 6th June, 2025

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Signature of Head of the Department

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# TABLE OF CONTENTS


| Serial Number | Topic | Page Number |
|---|---|---|
| 1. | List of Tables | 7 |
| 2. | List of Figures | 8 |
| 3. | Chapter 01: Abstract | 9 |
| 4. | Chapter 02: Keywords and Abbreviations | 9 |
| 5. | Chapter 03: Introduction | 10-14 |
| 6. | Chapter 04: Review of Literature | 14-16 |
| 7. | Chapter 05: Scientific Rationale of the Study | 16 |
| 8. | Chapter 06: Objectives | 17 |
| 9. | Chapter 07: Materials & Methodology<br>7.1 Preparation methods<br>7.1.1 Measurement of pH, Temperature, TDS, Conductivity and Microbiological Parameters<br>7.1.2 DO & BOD prep<br>7.1.3 Chloride & Salinity prep<br>7.1.4 Total alkalinity<br>7.1.5 Free Co2<br>7.1.6 Reactive phosphate prep<br>7.2 Methodology for water quality parameters<br>7.2.1 DO & BOD methodology<br>7.2.2 Chloride & Salinity methodology<br>7.2.3 Free CO2 methodology<br>7.2.4 Alkalinity methodology<br>7.2.5 Reactive phosphate methodology | 17-27 |
| 10 | Chapter 08: Results & Discussions<br>8.1- 8.14 Mean temp, Mean pH, TDS, Conductivity, DO, BOD, Free CO2, Chloride, Salinity, Alkalinity, Phosphate, TTC, TC, & Water quality index | 28-46 |
| 11 | Chapter 09: Conclusion | 46-47 |
| 12 | Chapter 10: Appendix | 48-62 |
| 13 | Chapter 11: References | 63-64 |
| 14 | Plagiarism Report | 65 |

# LIST OF TABLES


| Serial Number | Table Number | Title | Page Number |
|---|---|---|---|
| 1. | 10.1 | Monthly Variation of Mean Temp | 48 |
| 2. | 10.2 | Monthly Variation of Mean pH | 49 |
| 3. | 10.3 | Monthly Variation of TDS | 50 |
| 4. | 10.4 | Monthly Variation of Conductivity | 51 |
| 5. | 10.5 | Monthly Variation of DO | 52 |
| 6. | 10.6 | Monthly Variation of BOD | 53 |
| 7. | 10.7 | Monthly Variation of Free CO2 | 54 |
| 8. | 10.8 | Monthly Variation of Chloride | 55 |
| 9. | 10.9 | Monthly Variation of Salinity | 56 |
| 10. | 10.10 | Monthly Variation of Alkalinity | 57 |
| 11. | 10.11 | Monthly Variation of Phosphate | 58 |
| 12. | 10.12 | Monthly Variation of TTC | 59 |
| 13. | 10.13 | Monthly Variation of TC | 60 |

# LIST OF FIGURES


| Serial Number | Figure Number | Title | Page Number |
|---|---|---|---|
| 1. | 3.1 | Aquaculture bheri in East Kolkata Wetlands, with water spread and managed embankments | 11 |
| 2. | 3.3 | Canal in East Kolkata Wetlands receiving wastewater entering bheris | 13 |
| 3. | 7.2.1(a-c) | DO & BOD sample: (a) Collection, (b) After $MnSO_4$ & Alkali azide, (c) After conc.$H_2SO_4$ | 20-21 |
| 4. | 7.2.2 (a-c) | Chloride sample: (a) Collection, (b) With Potassium chromate, (c) After titration with $AgNO_3$ | 22-23 |
| 5. | 7.2.3 (a-b) | Free $CO_2$ sample: (a) Before titration, (b) With phenolphthalein, titrated with 0.01 N NaOH | 23-24 |
| 6. | 7.2.4 (a-c) | Alkalinity sample: (a) Without indicator, (b) With phenolphthalein & methyl orange indicator (c) After titration with $H_2SO_4$ | 25-26 |
| 7. | 8.1 | Monthly Mean Temperature (°C) across Sites and Canal | 28 |
| 8. | 8.2 | Monthly Mean pH across Sites and Canal | 29 |
| 9. | 8.3 | Monthly TDS (mg/L) across Sites and Canal | 31 |
| 10. | 8.4 | Monthly Conductivity (µS) across Sites and Canal | 32 |
| 11. | 8.5 | Monthly DO (mg/L) across Sites and Canal | 33 |
| 12. | 8.6 | Monthly BOD (mg/L) across Sites and Canal | 34 |
| 13. | 8.7 | Monthly Free $CO_2$ (mg/L) across Sites and canal | 36 |
| 14. | 8.8 | Monthly Chloride (mg/L) across Sites and Canal | 37 |
| 15. | 8.9 | Monthly Salinity (mg/L) across Sites and Canal | 38 |
| 16. | 8.10 | Monthly Alkalinity (mg/L) across Sites and Canal | 40 |
| 17. | 8.11 | Monthly Phosphate (mg/L) across Sites and Canal | 41 |
| 18. | 8.12 | Monthly TTC (CFU/mL) across Sites and Canal | 42 |
| 19. | 8.13 | Monthly TC (CFU/mL) across Sites and Canal | 44 |
| 20. | 8.14 | Spatial WQI variation in pond and canal waters during summer | 45 |
| 21. | 10.1 (a-b)<br>10.2 (a-b)<br>10.3 (a-b)<br>10.4 (a) | (a) MacConkey Agar-Site 1 (TC) (b) MacConkey Agar-Site 1 (TTC)<br>(a) MacConkey Agar-Site 2 (TC) (b) MacConkey Agar-Site 2 (TTC)<br>(a) MacConkey Agar-Site 3 (TC) (b) MacConkey Agar-Site 3 (TTC)<br>(a) MacConkey Agar-Canal (TC) | 61-62 |

# CHAPTER 01: ABSTRACT

The East Kolkata Wetlands (EKW) function as a natural wastewater treatment zone for the city, utilizing a single-pond system and are internationally acknowledged as a Ramsar site for their ecological importance. However, the specific mechanisms driving this natural purification process are still not well understood. Thus, to gain insight, efforts have been made in this study to understand the efficiency of the EKW pond in treating the sewage water and checking the parameter's spatiotemporal variations. For this purpose, different parameters such as DO, BOD, faecal coliforms, etc. have been measured monthly for six months from the pond and the adjacent canal, which serves as a primary inlet to the pond. The results demonstrated a progressive reduction in microbial load and a noticeable enhancement in water quality indicators was observed as the wastewater moved from canal as the water transitioned from the canal and passed through the aquaculture-based pond system locally known as the Bheri. It also depicted the role of agricultural runoff during monsoon and increased organic pollution in summer. Thus, the study provided an insight into the condition of the pond water, justifying the importance of the traditional method of fish farming in the EKW aquaculture facility. Furthermore, the observed fluctuations in water quality not only reflect the system's interaction with seasonal cycles but also emphasize the need for continuous assessment of faecal contamination. The presence of coliform bacteria serves as a key microbial indicator for sanitary risk, making their monitoring essential for both environmental and public health. The sustainable design of the bheri system showcases the fusion of ecological engineering and traditional practices, making it an effective low-cost model for wastewater reuse. This study, therefore, contributes to a better understanding of the ecological functions of EKW and supports its preservation as a vital urban wetland offering ecosystem services and livelihood security.

# CHAPTER 02: KEYWORDS & ABBREVIATIONS

## 2.1. Keywords

East Kolkata Wetland, Spatiotemporal, Bheri, Aquaculture, Microbial load, Faecal Coliform, Water Quality, Biological Oxygen Demand (BOD).

## 2.2. Abbreviations

EKW- East Kolkata Wetland; BOD- Biological Oxygen Demand; DO- Dissolved Oxygen; COD- Chemical Oxygen Demand; FC- Faecal Coliform; WQI- Water Quality Index; APHA- American Public Health Association; TDS- Total Dissolved Solids; SD- Standard Deviation; WWT- Wastewater Treatment; TTC- Thermotolerant Coliform; TC- Total Coliform

# CHAPTER 03: INTRODUCTION

## 3.1 Aquaculture Bheri Of East Kolkata Wetland :

Aquaculture has seen continuous growth over the past hundred years, becoming a vital approach to meeting the increasing global need for food, better nutrition, employment and economic progress. In the past, many communities relied heavily on capturing wild fish, but this method became unsustainable as fish populations declined due to factors like overfishing, environmental pollution, and long-term climate variability (Naylor et al. 2000).

To response to these issues, the “Blue Revolution” emerged in the late 20th century—similar in spirit to the earlier Green Revolution in farming. In India, aquaculture has rapidly expanded under the influence of the Blue Revolution, emerging as one of the nation's most dynamic and fastest-growing food production sectors. A key contributor to this growth is the presence of distinctive regions where fish farming is closely integrated with local cultural practices and natural ecosystems (Lakra et al. 2021).

One such region is the East Kolkata Wetlands (EKW), renowned for their distinctive aquaculture system that operates using treated urban wastewater. Located along the eastern edge of Kolkata, this system transforms urban wastewater into a productive resource through an interconnected network of fish ponds known locally as "Bheris." These shallow ponds not only support fish farming but also help in reducing the environmental impact of municipal wastewater (Bunting et al. 2010; Mandal et al. 2023).

A Bheri is a shallow pond, typically ranging between 15 to 34 hectares, that receives wastewater from city drainage through an interconnected canal system. These ponds are carefully designed and managed to encourage the settling of suspended solids and the absorption of nutrients by phytoplankton and aquatic plants—creating ideal conditions for fish farming. The EKW’s Bheri system represents an effective fusion of indigenous practices and ecological design, offering a natural method for treating wastewater while simultaneously supporting aquaculture (Mandal et al. 2023).

This sustainable model not only alleviates the pollution burden on nearby water bodies but also forms the economic backbone for thousands of local residents engaged in pisciculture. One of the key features of EKW is the aquaculture bheris, used for fish and other aquatic species cultivation. However, the water quality and faecal coliform dynamics in these bheris have become a growing concern. The system’s productivity, however, is heavily dependent on the quality of water flowing through it. Therefore, continuous monitoring and maintenance of water standards are vital—not only to safeguard public health but also to maintain the ecological stability and functionality of this unique urban wetland ecosystem (Hussan et al. 2019).

To further enhance the sustainability of the aquaculture system, current research emphasizes the importance of probiotics in maintaining water quality and fish well-being. These beneficial

micro-organisms help regulate microbial populations in the bheri ponds, suppress harmful pathogens, and enhance the recycling of nutrients. The adoption of probiotics within the East Kolkata Wetland reflects a successful blend of traditional practices with modern biological advancements, ensuring the long-term health and productivity of the wetland's aquaculture network (Sona et al. 2021).

**FIGURE 3.1**- View of an aquaculture bheri (pond) in the East Kolkata Wetlands, showing extensive water spread and managed embankments used for traditional fish culture

## 3.2 East Kolkata Wetland: An Integrated Aquaculture System

The East Kolkata Wetlands designated as Ramsar site since 2002, span over 12,500 hectares on the Eastern edges of Kolkata. They are recognized globally for their unique use of wastewater in agriculture and aquaculture (Saha et al. 2021). The East Kolkata Wetlands consist of a diverse mix of water channels, farmland, rice paddies, waste-fed fisheries, and swampy areas (Mukherjee et al., 2016). East Kolkata Wetland exemplifies how a natural ecosystem can be adapted to meet urban challenges- particularly the management of domestic wastewater while maintaining ecological functions (Bhattacharya et al., 2019). These wetlands serve a dual function: They act as natural waste assimilation zones for urban sewage and simultaneously sustain a large-scale, resource-efficient aquaculture system (Rath et al., 2018).The EKW serves multiple roles- as a food production centre, a natural water treatment plant, a biodiversity hotspot, and a climate regulator (Das et al., 2020). The ecosystem is home to various fish species, migratory birds, and native flora (Saha & Saha, 2020). Its ecological services provide significant savings to the municipal corporation in terms of water treatment

costs and support the livelihoods of over 50,000 people engaged in fisheries and farming (Hussain et al., 2017). The inhabitants of the wetlands, predominantly from low-income fishing and farming communities, have over decades developed sustainable practices that transform waste into wealth (Chakraborty et al., 2019). Apart from fish production, the nutrient-rich sludge and water support agriculture in surrounding lands, especially the cultivation of rice and vegetables (Mandal et al., 2020). The resilience of this system lies in its adaptability: it absorbs pollution loads while remaining productive (Basu et al., 2018). Nonetheless, the East Kolkata Wetlands are facing significant stress due to fast-paced urban growth, shifting land-use practices, and rising population density, all of which are disrupting the area's fragile ecological balance (Nair et al., 2021). One of the growing concerns in recent years is the deteriorating water quality, especially microbial contamination, which directly affects aquaculture and public health (Sen & Ghosh, 2020).

## 3.3 Water Quality of East Kolkata Wetland

The Water quality of the East Kolkata Wetlands (EKW), a Ramsar-designated site, is a function of both its natural wetland processes and anthropogenic inputs. It influences biodiversity, fish productivity, and public health, making its analysis an essential part of wetland conservation and management strategies. Among the most significant contributors are the volume and nature of incoming sewage, the hydrodynamics of the bheris (traditional fish ponds), and seasonal variations in climatic conditions, particularly temperature and rainfall (Saha et al., 2021). While the inflow of wastewater provides some nutrients, it also introduces pollutants- chemical, physical, and biological. Tracking water quality is essential for evaluating the sustainability and safety of the East Kolkata Wetlands (EKW), particularly in relation to its aquaculture operations (Bunting et al., 2010). In the context of the EKW region, several critical water quality parameters must be monitored closely such as-

- **Physical Parameters**: Temperature, pH, TDS, and Conductivity
- **Chemical Parameters**: Biological oxygen demand (BOD), Dissolved oxygen (DO), Phosphate, Free $CO_2$, Alkalinity, Chloride, and Salinity
- **Biological Parameters**: Total coliforms (TC), and Thermotolerant coliforms (TTC)

These parameters interact with each other to shape the overall ecological balance of the wetlands (Mandal et al., 2023).

Among these, BOD and DO levels are critical indicators of organic pollution and the system's oxygen balance. High BOD indicates high organic content, which, if not matched by adequate oxygenation, leads to hypoxia- affecting both microbial dynamics and fish health. Nutrient levels (especially nitrogen and phosphorus) influence phytoplankton growth, which in turn supports fish productivity. However, nutrient enrichment can also trigger eutrophication, which lead to algal blooms, oxygen depletion, and fish death rate (Chatterjee et al., 2011).

Seasonal variations play a major role in shaping water quality. During the monsoon, inflows increase and dilution occurs, while in summer, lower water levels and higher temperatures can exacerbate pollution effects. Spatially, water quality tends to degrade closer to the wastewater inlet points and improves toward the outlet, due to natural sedimentation and microbial

degradation processes. Yet, despite this self-purifying, the cumulative effects of unchecked waste inflow are evident. One of the most persistent and under-addressed issues in this regard is microbial contamination- particularly the presence of faecal coliform bacteria (Hussan et al., 2019).

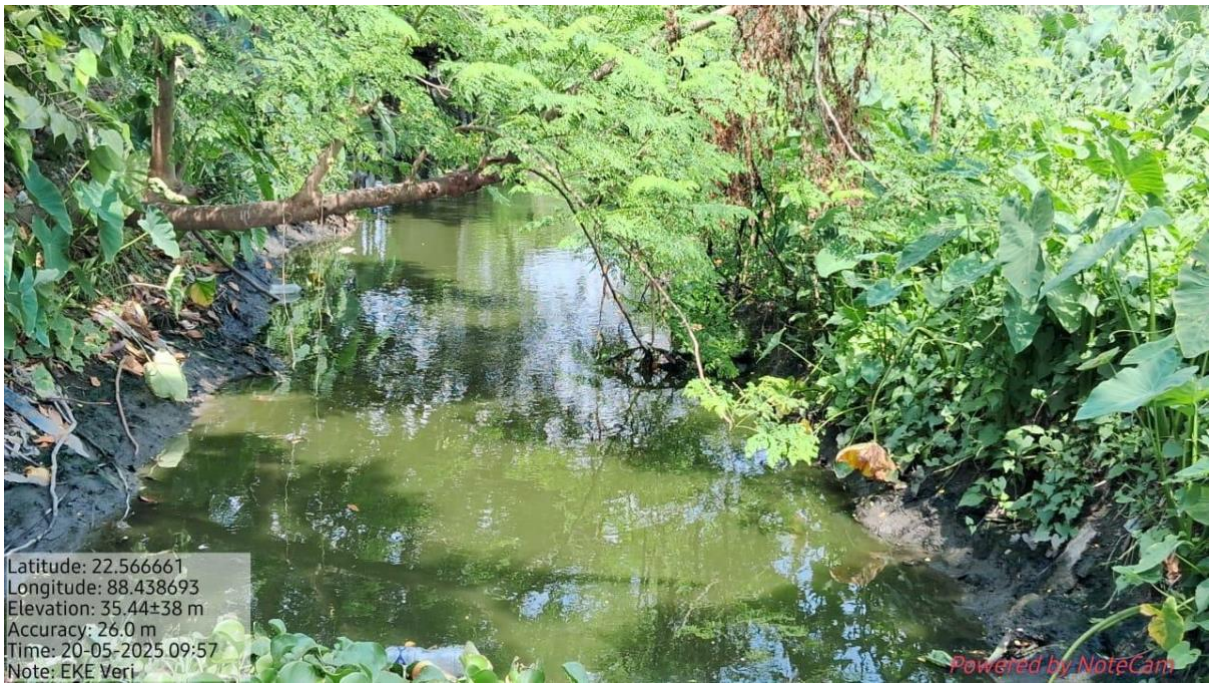


**FIGURE 3.3**- A canal segment within the East Kolkata Wetlands receiving wastewater, which later enters the aquaculture bheris, influencing water quality

## 3.4 Faecal Coliform Dynamics in Aquaculture Bheri of East Kolkata Wetland

Faecal coliforms are a subset of coliform bacteria that originate specifically from the faeces of warm-blooded animals, including humans. The most well-known species among them is Escherichia coli (E.coli), which is used as an indicator organism for faecal contamination. The detection of faecal coliforms in water suggests the possible presence of enteric pathogens like Salmonella, Shigella, Vibrio cholerae, and various viruses and protozoa that can pose severe health risks. In the bheris of EKW, faecal coliforms enter the system primarily through untreated or partially treated domestic wastewater. Studies conducted in the EKW have reported faecal coliform concentrations often exceeding national and international limits for aquaculture waters (Raychaudhuri et al., 2008). The dynamics of faecal coliform populations within the EKW are complex and closely linked to the broader patterns of wastewater inflow, environmental conditions, and aquaculture practices. Faecal coliforms serve as bioindicators of ecosystem degradation. Their abundance often correlates with high BOD, low DO, and other

signs of organic overload. Monitoring their dynamics thus provides a window into both the health of the wetland ecosystem and the safety of its aquaculture output (Saha et al., 2021).

Faecal coliform dynamics affect the aquaculture bheris of the East Kolkata Wetlands (EKW) in many ways which impact water quality, fish health, productivity, and public health. Here's a breakdown of how these bacteria influence the system:

1. **Water Quality Degradation**-

   High concentration of faecal coliform in bheris implies the presence of pathogens and a high organic load, which can-

- Increase biological oxygen demand (BOD), reducing dissolved oxygen (DO) essential for fish survival
- Lead to eutrophication due to nutrient overloading, causing algal blooms that further deplete oxygen
- Contribute to the formation of toxic substances like ammonia and hydrogen sulfide through anaerobic decomposition (Mandal et al., 2023).

2. **Bheri Ecology Disruption**-

- Faecal matter supports the growth of unwanted microbial communities and can shift the natural microbial balance, affecting the decomposition processes and nutrient cycles (Chatterjee et al., 2011).

# CHAPTER 04: REVIEW OF LITERATURE

The East Kolkata Wetlands (EKW), a Ramsar site of international importance, is renowned for its unique and sustainable use of wastewater for aquaculture. This urban wetland system not only supports biodiversity but also plays a crucial role in urban wastewater recycling and livelihood support for local communities (Ghosh, 2005). However, with growing urbanization and discharge of untreated or partially treated sewage, the water quality in these bheris is increasingly under threat, particularly with reference to microbial contamination.

## 4.1 Water Quality in Aquaculture Systems

Aquaculture practices in wetlands such as the EKW depend heavily on water quality parameters like dissolved oxygen (DO), biological oxygen demand (BOD), chemical oxygen demand (COD), pH, temperature, ammonia, and nutrient concentrations (Boyd, 1990). Regular influx of sewage in the bheris provides organic matter and nutrients beneficial for plankton growth,

thus enhancing fish productivity. However, excess nutrient loading can lead to eutrophication, hypoxia, and altered microbial dynamics (Kumar et al., 2004). Studies by Mukherjee et al. (2012) have documented seasonal variations in DO and BOD in bheri systems, correlating these with algal bloom occurrences and fish mortality.

## 4.2 Faecal Coliform as an Indicator of Microbial Pollution

Faecal coliforms, especially *Escherichia coli*, are widely recognized as reliable indicators of faecal contamination in aquatic environments. Their presence suggests possible intrusion of enteric pathogens that pose serious risks to human health, especially in fish-handling communities and downstream users (WHO, 2006). In the context of EKW, primary sources of faecal coliforms include domestic sewage, urban runoff, and occasional livestock activity near water bodies (Niyogi & Mitra, 1999).

Das and Acharya (2003) observed significantly high faecal coliform levels in several bheris of EKW, particularly during monsoon due to increased surface runoff and sewage inflow. The bacterial load showed diurnal and seasonal variations, influenced by sunlight (UV inactivation), temperature, and organic matter content. A study by Banerjee et al. (2015) linked coliform counts with fish health, reporting increased disease incidence and reduced marketability of fish during peak contamination months.

## 4.3 Dynamics and Control Measures

The microbial dynamics in bheris are influenced by a complex interplay of environmental, hydrological, and anthropogenic factors. Seasonal trends often show higher coliform counts during monsoon and post-monsoon seasons (Roy et al., 2006). High temperatures in summer can lead to faster bacterial die-off but may also enhance nutrient cycling, which indirectly supports microbial growth. Sediment also acts as a reservoir for faecal bacteria, releasing them back into the water column under disturbances.

Efforts to control faecal contamination include proper wastewater management, constructed wetlands, sediment dredging, and promoting fish species resistant to microbial diseases (Mitra et al., 2008). Ecohydrological approaches, such as controlled water flow and maintenance of vegetation buffer zones, have shown promise in reducing pathogen load.

## 4.4 Research Gaps and Future Directions

Although several studies have assessed physico-chemical parameters and general microbial quality of EKW water (Mukherjee et al., 2012; Mitra et al.,2008), limited research exists on the fine-scale temporal dynamics of faecal coliforms in relation to aquaculture productivity. While Banerjee et al. (2015) and Das & Acharya (2003) have examined coliform levels, there

is a lack of integrated approaches combining microbial analysis with spatial mapping, water chemistry, and fish health assessments. Moreover, existing literature tends to focus either on environmental quality or aquaculture yield, often treating them as separate domains rather than interconnected systems (Kumar et al., 2004; Roy et al., 2006). This highlights the need for interdisciplinary studies that can evaluate microbial risks and water quality within the operational context of wetland-based aquaculture systems like those in EKW.

# CHAPTER 05: Scientific Rationale of the Study

The East Kolkata Wetlands (EKW), a designated Ramsar site, represent a unique ecological and socio-economic system where traditional aquaculture practices coexist with urban wastewater reuse. These wetlands serve as natural biofilters by receiving and partially treating the domestic sewage of Kolkata, which is then channeled into aquaculture ponds or bheries. While this model supports nutrient recycling and fish production, it also poses significant risks to aquatic health and public safety due to potential microbiological contamination, especially from faecal coliforms.

Faecal coliforms are widely recognized as key indicators of microbial water quality and are used to assess the potential presence of pathogenic organisms originating from human or animal waste. Elevated faecal coliform levels not only compromise aquatic biodiversity and fish health but also raise serious concerns regarding the safety of fish products consumed by local populations. In aquaculture systems such as those in EKW, where untreated or partially treated sewage is used as a nutrient source, the dynamics of faecal coliform populations are influenced by a complex interplay of environmental parameters such as temperature, dissolved oxygen, nutrient concentrations, and hydrological conditions.

Despite the ecological and economic importance of EKW, there remains a notable gap in the systematic assessment of water quality and the spatiotemporal behaviour of faecal coliforms in its aquaculture units. Understanding how seasonal variations and water quality parameters influence microbial contamination is critical for developing evidence-based management practices. Moreover, as climate variability and anthropogenic pressures continue to impact wetland ecosystems, monitoring faecal contamination trends becomes increasingly essential to protect public health and preserve the ecological integrity of the wetlands.

Therefore, this study aims to provide a comprehensive evaluation of the physico-chemical characteristics and faecal coliform dynamics across two representative bheries of EKW. By correlating microbiological data with environmental parameters over different seasons, the research seeks to uncover patterns and drivers of contamination. The outcomes will contribute to sustainable aquaculture management, promote safer reuse of wastewater in fish farming, and support informed policy-making for wetland conservation.

# CHAPTER 06: OBJECTIVES

1. To evaluate the Water Quality Index (WQI) of the aquaculture bheris in East Kolkata Wetland using physico-chemical parameters, in order to assess the suitability of water for aquaculture practices.

2. To analyze the spatio-temporal variation in key water quality parameters namely pH, Tds, Conductivity, Temp, BOD, DO, Free CO2, Phosphate, Chloride, alkalinity.

3. To assess the spatial distribution of faecal coliform counts to understand the extent of microbial contamination in the wetland water.

# CHAPTER 07: MATERIALS & METHODOLOGY

## 7.1 Preparation Methods

**7.1.1** **pH**, **Temperature, TDS, Conductivity** would be measured using respective meters. For microbiological parameters, 2ml of the water sample would be taken, centrifuged at 13rpm for 13min and resuspended in 1ml distilled water thereby through spread plate technique 100 microL of water would be spread on Mac Conkey (**TTC** incubation 44°C overnight and **TC** 37°°C overnight).

**7.1.2** **DO and BOD prep** APHA 1976 (14th ed.)

- Manganous sulfate solution: For 50ml $MnSO_4.H_2O$ solution, 18.2 gm $MnSO_4$ was weighted and dissolved in 50ml distilled water.
- Alkaline - iodide - azide reagent: For 50 ml Alkaline - iodide - azide solution, 25 gm NaOH (Sodium Hydroxide), 7.5 gm KI (Potassium iodide) and 0.5 gm $NaN_3$ (Sodium Azide) were weighted and dissolved in 50 ml distilled water.
- Conc. $H_2SO_4$ (Sulfuric acid) (98%)
- Sodium thiosulfate solution: For 250 ml $Na_2S_2O_3$ solution, 1.551 gm of $Na_2S_2O_3$, $5H_2O$ was weighted and dissolved in 250 ml distilled water.
- Potassium Dichromate solution: For 250 ml 0.025 (N) $K_2Cr_2O_7$ solution, exactly 0.3064 gm $K_2Cr_2O_7$ was weighted and dissolved in a 250 ml volumetric flask with little amount of distilled water and then volume make up was done up to the 250 ml mark of that flask with distilled water.

- Starch indicator solution: 1 gm of starch was weighted and dissolved in 10ml distilled water and pour it into 100ml boiling distilled water and mix well. If any precipitate formed, transferred the supernatant into a flask and use it as starch indicator.

### 7.1.3 Chlorides/ Chlorinity/ Salinity prep APHA 1976 (14th ed.)

- Silver nitrate solution [~0.0141 (N)] : For 250 ml 0.0141 (N) $AgNO_3$ solution, 0.599 gm of $AgNO_3$was weighted and dissolved in 250 ml volumetric flask with distilled water.
- Sodium Chloride Solution [0.0141 (N)] : For 250 ml 0.0141 (N) NaCl solution, exactly 0.206 gm of NaCl was weighted and dissolved in 250 ml volumetric flask and the volume was made up to 250 ml mark of the flask with distilled water.
- Potassium Chromate indicator solution: For 50 ml K2CrO4 solution, 2.5 gm of $K_2CrO_4$ was weighted and dissolved in 50 ml of distilled water.

### 7.1.4 Total alkalinity

- Sulphuric Acid Solution (0.02N): Take 0.1388 ml of Conc. Sulphuric acid in 250 ml of distilled $H_2O$.
- Phenolphthalein indicator 0.5 gm of Phenolphthalein was weighted and dissolved in 50ml of 50% ethyl alcohol solution.
- Methyl Orange Indicator Solution - 0.4 g/l. Dissolve 0.1 g of methyl orange in 50 ml of rectified spirit and dilute to 250 ml with water.

### 7.1.5 Free Co2 (APHA 2017)

- Sodium hydroxide solution - 0.01N NaOH solution was prepared by mixing 400mg NaOH in distilled water and volume was made up to 1litre.
- Phenolphthalein indicator solution: 0.5 gm of Phenolphthalein was weighted and dissolved in 50ml of 50% ethyl alcohol solution.

### 7.1.6 Reactive Phosphate prep APHA 1976 (14th ed.)

- Stock Phosphate Solution ($KH_2PO_4$) [250 µg/ml$PO^{3-}_4$]: For 100 ml stock phosphate solution, 0.1097 gm Potassium dihydrogen orthophosphate ($KH_2PO_4$) was weighted and dissolved in 100 ml distilled water.
- Ascorbic Acid: For ascorbic acid solution, 1.76 gm ascorbic acid was weighted and dissolved in 100 ml distilled water.
- Ammonium Molybdate solution: For ammonium molybdate solution, 4 gm Ammonium heptamolybdate tetrahydrate was weighted and dissolved in 100 ml distilled water.

- Potassium Antimonyl Tartrate solution: 1.3715 gm Potassium Antimony oxide tartrate hemihydrate was weighted and dissolved in 500 ml distilled water.
- Sulfuric Acid [5 (N)]: 34.72 ml Conc. $H_2SO_4$ was measured and volume was made upto 250 ml with distilled water.
- Phenolphthalein indicator solution: 0.5 gm of Phenolphthalein was weighted and dissolved in 50ml of 50% ethyl alcohol solution.
- Combined Reagent: For 50 ml combined reagent, mixed the above reagents in the following proportion: 25 ml 5(N) $H_2SO_4$, 2.5 ml potassium antimonyl tartrate solution, 7.5 ml ammonium molybdate solution and 15 ml ascorbic acid. These reagents were mixed well after addition of each reagent.

## 7.2 Methodology for Water Quality Parameters

### 7.2.1 DO and BOD methodology

BOD was measured using Direct method. For this method sample was collected in two separate bottles at the same time and same place. Then one of them was initially fixed with 2ml of $MnSO_4$ and 2 ml of alkali azide iodide reagent and mixed well. Then the other bottle was placed in incubator at 37℃ for 5 days. After 30 minutes, the fixed sample was acidified by adding 2 ml Conc. $H_2SO_4$ to completely dissolve the precipitation. The other sample which was placed in the incubator was fixed after 5 days and then acidified and mixed well. After acidification, Dissolved oxygen (DO) was measured from both the samples and BOD was calculated by this formula: $DO_{initial}$-$DO_{final}$ = BOD.

So the protocol by which DO was calculated is as followed:

- After the sample water was fixed with alkali azide iodide reagent (alkaline KI) and $MnSO_4$, a brownish precipitate was formed which was dissolved by adding conc. $H_2SO_4$. Then a dark brownish-orange coloured solution formed.
- Sodium thiosulphate ($Na_2S_2O_3$) was taken as titre solution in burette and after volume correction 50ml of sample was taken as titrate. Titration was done till the solution turned straw yellow in colour.
- 2-3 drops of starch indicator solution was added to make the titrate dark blue. The resulting solution titrated again till it turned colourless.

The experiment was done three times and the mean was considered as the final value.

**FIGURE 7.2.1 (a)- Water sample collected for DO and BOD analysis**

**FIGURE 7.2.1 (b)- After adding $MnSO_4$ and Alkali azide**

**FIGURE 7.2.1 (c)- After adding Conc.$H_2SO_4$**

### 7.2.2 Chlorides/ Chlorinity/ Salinity methodology

- 20 ml of sample water was taken in a conical flask and 2-3 drops of potassium chromate ($K_2CrO_4$) indicator were added to the sample. The solution turned yellow in colour.
- Then the sample water was titrated against Silver nitrate ($AgNO_3$) solution until the colour of the solution changed to Reddish-yellow in colour.
  The experiment was repeated three times and the mean value was considered as final value.

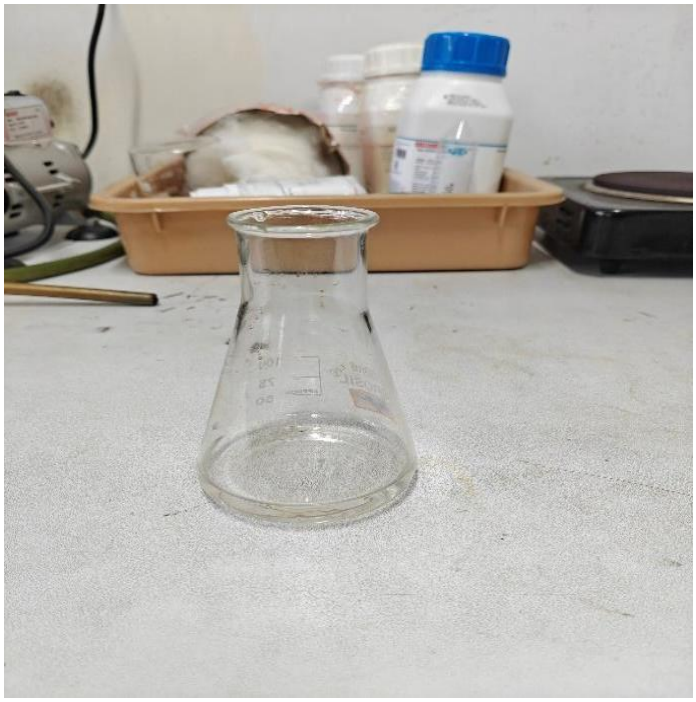

**FIGURE 7.2.2 (a)- Water sample for Chloride titration**

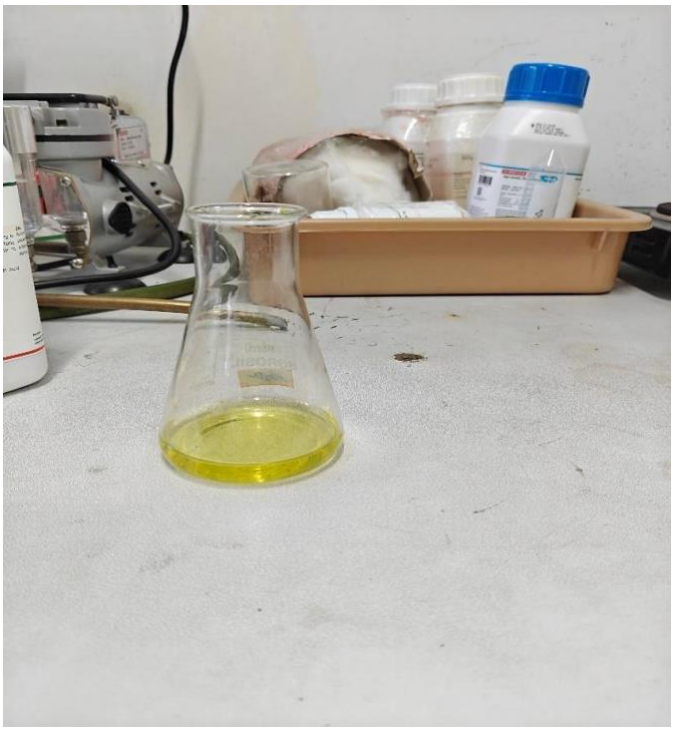

**FIGURE 7.2.2 (b)- After adding indicator Potassium Chromate**

**FIGURE 7.2.2 (c)- After titration against Silver Nitrate**

### 7.2.3 Free $CO_2$ methodology

Free $CO_2$ was measured by titrimetric method for free carbon-di-oxide estimation as described in APHA, 2017. The results were expressed in mg/l.

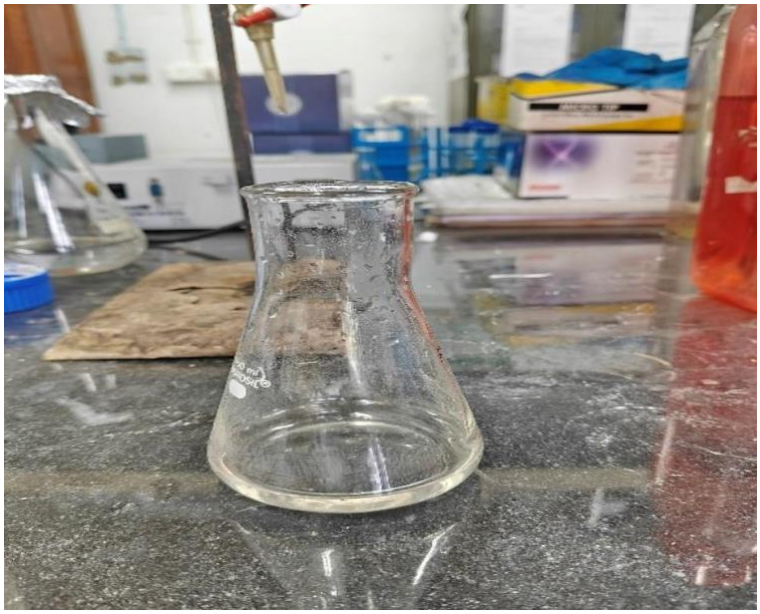

**FIGURE 7.2.3 (a)- Water sample in conical flask before titration for Free $CO_2$ estimation**

**Reagents prep**

- Sodium hydroxide solution - 0.01N NaOH solution was prepared by mixing 400mg NaOH in distilled water and volume was made up to 1litre.
- Phenolphthalein indicator.

**Procedure**

- 20ml of the sample were taken in a 50ml volumetric flask and 2-3drops of Phenolphthalein indicator was added and mixed well. The mixture was titrated against 0.01N NaOH solution. Titration was continued until the appearance of faint pink colour indicating pH 8.3

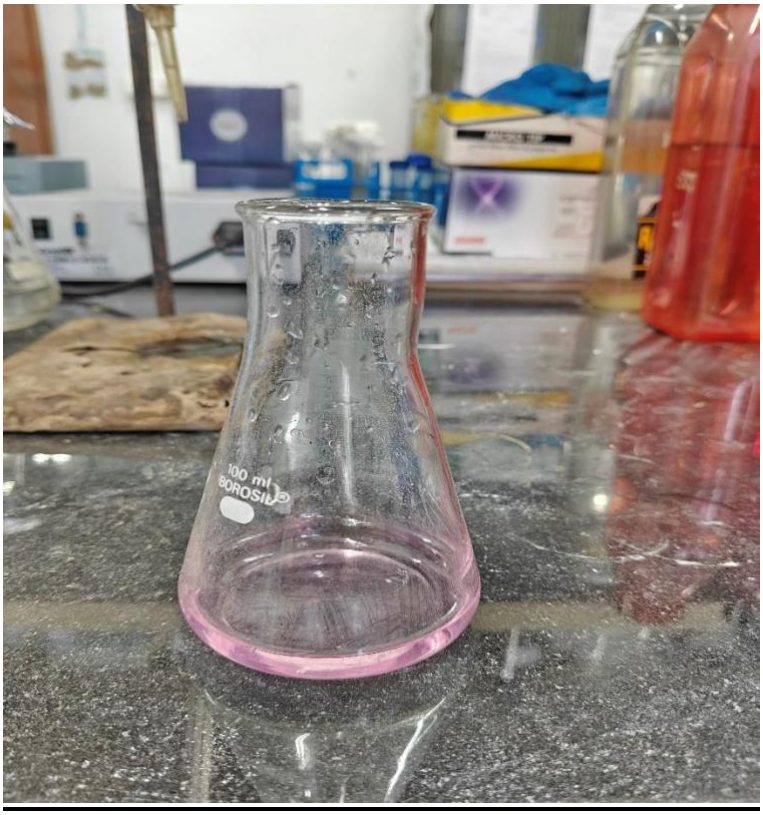


**FIGURE 7.2.3 (b)- Sample was added with phenolphthalein and the mixture was titrated against 0.01 N NaOH**

### 7.2.4 Alkalinity methodology

- Sample (20ml) was taken in a conical flask and addition of 3-4 drops of phenolphthalein indicator to check if the colour of the solution turned pink.
- If it turned pink, the solution was titrated against N/50 $H_2SO_4$ till the solution again turned colourless.
- If the sample did not turn pink, or, after it turned colourless, 3-4 drops of methyl orange was added to turn the solution yellow.
- The solution was then titrated against N/50 $H_2SO_4$ till orange coloured solution appeared.
- The test was repeated thrice and the mean reading was taken as the final value.

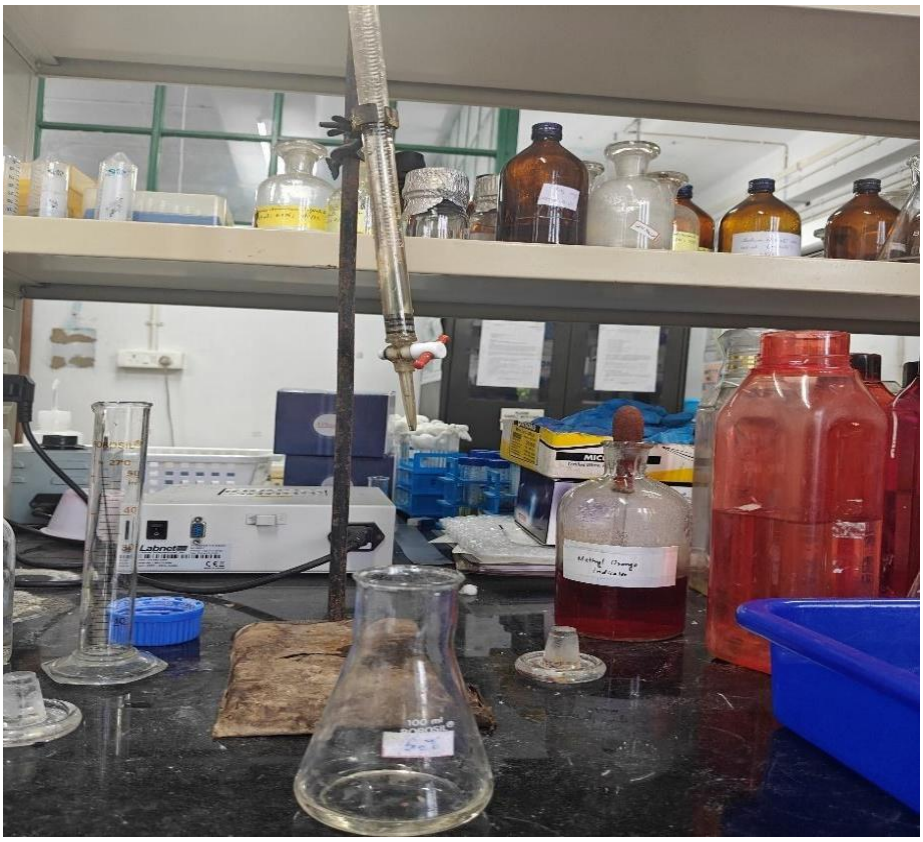

**FIGURE 7.2.4 (a)- Water sample for Alkalinity without any indicator**

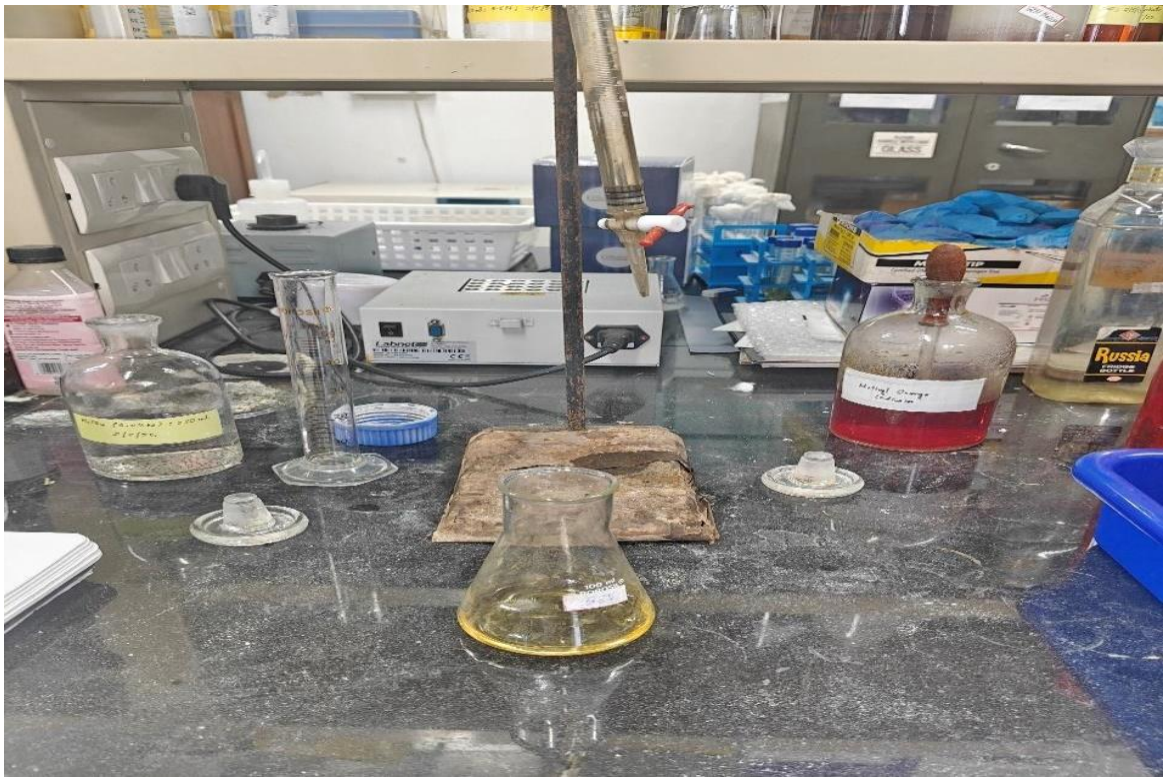


**FIGURE 7.2.4 (b)- Sample with phenolphthalein and methyl orange indicator**

**FIGURE 7.2.4 (c)- Colour change after titrated against 0.02 N $H_2SO_4$**

### 7.2.5 Reactive Phosphate methodology

✓ **Standard blank solution:**

- 4 ml of distilled water was taken in a 100 ml volumetric flask then few drops of sulphuric acid and 8 ml of combined reagent were added. Finally volume was made up to 100 ml by distilled water.

✓ **Preparation of Standard solution for Reactive Phosphate (for standard curve) (10ug/ml):**

- 4 ml of stock solution ($KH_2PO_4$) (250µg/ml) was taken in 100 ml volumetric flask. Then 4 ml distilled water and few drops of Phenolphthalein were added.
- If the solution turned red then 5 (N) $H_2SO_4$ was added drop wise until the solution became colourless.
- Volume was made up to 100 ml by distilled water and the phosphate concentration standard solution became 10µg/ml.
- Then 0.125 ml, 0.25 ml, 0.5 ml, 1 ml, 2 ml, 3 ml and 4 ml standard reactive phosphate solution was taken in 7 volumetric flasks respectively.
- 8 ml combined reagent was added in each flask and volume made up to 100 ml by distilled water. Then it was kept for 10 minutes to complete the reaction.
- Spectrophotometric Analysis
- Absorbance was set up to zero (0) using blank solution. Then the absorbances of the above mentioned standard solution of different concentrations were measured one by one against blank solution at 880 nm. All the readings were recorded and the standard curve was plotted.
- Reactive phosphate for the sample -
- 50 ml of filtered sample water was taken in a volumetric flask. Then few drops of Phenolphthalein were added.
- If the solution turned red then 5 (N) $H_2SO_4$ was added drop wise until the solution became colourless.
- 8 ml of combined reagent was added and volume was made up to 100 ml and kept for 10 minutes for the blue colour development.
- After the blue colour development, absorbance was measured at 880 nm against the standard blank solution and the reading was recorded.

  Finally, from the standard curve we could extrapolate the phosphate concentration corresponding to the absorbance.

# CHAPTER 08: RESULTS AND DISCUSSION

## 8.1 Mean Temperature

This section presents and interprets the physicochemical and microbial parameters measured across different sites and sampling months in the aquaculture bheris and canal of the East Kolkata Wetlands. Emphasis is laid on temperature variations, as temperature is a key ecological factor influencing aquatic productivity and fish health.

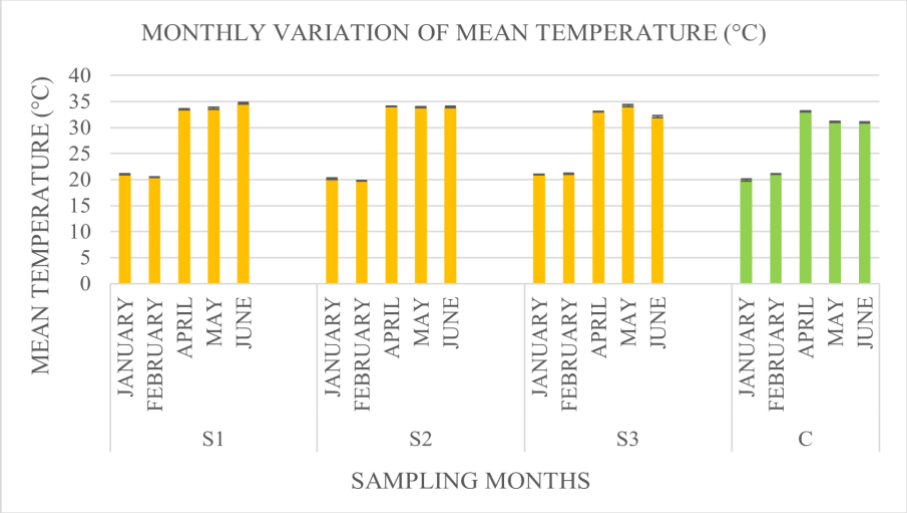


**FIGURE 8.1- Monthly Variation of Mean Temperature (°C) across Sampling Sites and Canal**

**Significance of Mean Temperature (°C) in the study-**

The mean temperature, ranging between 20°C and 35°C across all sites and months, holds significant ecological and aquacultural relevance in the East Kolkata Wetland's bheri system. This temperature range is critical because it directly influences metabolic activity, growth rate, and reproductive efficiency of fish species commonly cultivated in these wetlands. It influences the solubility of dissolved oxygen and regulates nutrient cycling, which are essential for maintaining water quality. Additionally, temperature exerts a strong influence on microbial behaviour, including the activity and survival of faecal coliform bacteria, which are key indicators of microbial contamination.

**Monthly Variation of Mean Temperature-**

The mean temperature recorded across all sampling locations- S1, S2, S3, and the Canal (C)- ranged from 20°C to 35°C between Jan and June. A clear seasonal trend emerged: lower temperatures during January and February, followed by a steady rise peaking in April to June, reflecting the regional climatic shift from winter to summer, where increased atmospheric

temperatures during the summer months elevate water temperatures correspondingly. At site S1, temperatures rose from just above 20°C in Jan-Feb to nearly 35°C by April-June. Similar thermal patterns were observed at S2 and S3, where winter months recorded lower thermal values, rising steadily during pre-monsoon period. The Canal site also reflected this seasonal rise, with temp values closely mirroring those of the bheri stations during the warmer months. The observed temperature range (20-35°C) falls within the permissible thermal limit for warm-water fish species recommended by CIFRI. While, lower winter temperatures may suppress fish metabolism and growth, elevated summer temperatures promote faster nutrient uptake and growth. However, temperatures nearing 35°C may lead to thermal stress, oxygen depletion, and increased disease risk. Therefore, seasonal thermal monitoring is critical for sustaining aquaculture efficiency in this ecosystem.

## 8.2 Mean pH

This section presents and interprets the monthly mean pH values observed across the selected aquaculture bheris and the canal in the EKW. Emphasis is placed on pH as a crucial physicochemical parameter that affects water quality, aquatic organism health, and overall productivity in aquaculture systems. pH influences various biological processes such as enzyme activity, nutrient availability, and microbial dynamics, making it essential to monitor in managed aquatic environments.

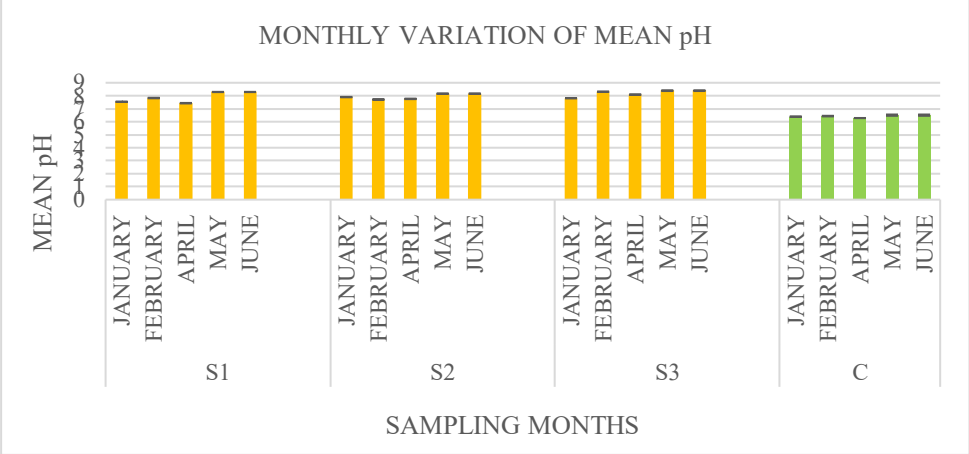


**FIGURE 8.2- Monthly Variation of Mean pH across Sampling Sites and Canal**

**Significance of Mean pH in the study-**

In aquaculture, maintaining an optimal pH range is vital for the health and productivity of aquatic organisms. pH directly impacts the solubility and bioavailability of nutrients and toxic substances in water. For freshwater fish culture, the recommended pH range generally lies between 6.5 and 8.5. Deviations outside this range can lead to physiological stress, reduced immunity, inhibited growth, and in severe cases, mortality among cultured species. Hence,

stable pH conditions are integral to maintaining water quality and ensuring fish welfare in aquaculture systems.

**Monthly Variation of Mean pH-**

The analysis of monthly mean pH values from January to June across three aquaculture sites (S1, S2, and S3) and one canal site (C) revealed relatively consistent trends throughout the study period. At all three aquaculture stations, the mean pH ranged between 7.4 and 8.4, demonstrating minimal fluctuation across the months. These values indicate a neutral to slightly alkaline environment, which is considered suitable for aquaculture practices. In contrast, the canal site consistently exhibited lower pH values, ranging between 6.8 and 7.2 during the same period. This comparatively lower pH suggests mildly acidic conditions in the canal water, which may be influenced by a higher organic load, and lack of natural buffering capacity. Although these values remain within a tolerable range for aquatic organisms, they are closer to the lower threshold of acceptability for aquaculture use. Maintaining appropriate pH levels is essential in aquaculture, as it affects physiological processes in fish, including respiration, excretion, and overall metabolic activity. The optimal pH range for most freshwater fish lies between 7.0 and 8.5. Values falling outside this range can impair enzymatic activity, reduce feed efficiency, and increase susceptibility to diseases. According to the BIS and the CPCB guidelines, the recommended pH range for surface waters suitable for fisheries is 6.5 to 8.5. All three bheri sites recorded values well within this permissible range, confirming favorable water quality for fish culture. The relatively stable pH in the bheris may be attributed to regular water exchange, and the buffering effect of dissolved salts and alkalinity, which help maintain the equilibrium. On the other hand, the canal's acidic tendency may result from limited water circulation, accumulation of decomposed organic matter, or external pollutant inflow, which can lower the buffering capacity and consequently reduce pH.

## 8.3 TDS (Total Dissolved Solids)

This section analyses the spatiotemporal variation of Total Dissolved Solids (TDS) across different aquaculture sites (S1, S2, S3) and the adjacent canal (C) in the East Kolkata Wetlands. TDS refers to the total concentration of dissolved substances such as salts, minerals, and organic matter present in water. TDS is an essential physicochemical parameter that reflects the concentration of dissolved salts, minerals, and organic matter present in water. It significantly influences aquatic productivity, osmoregulation in fish, and overall water quality.

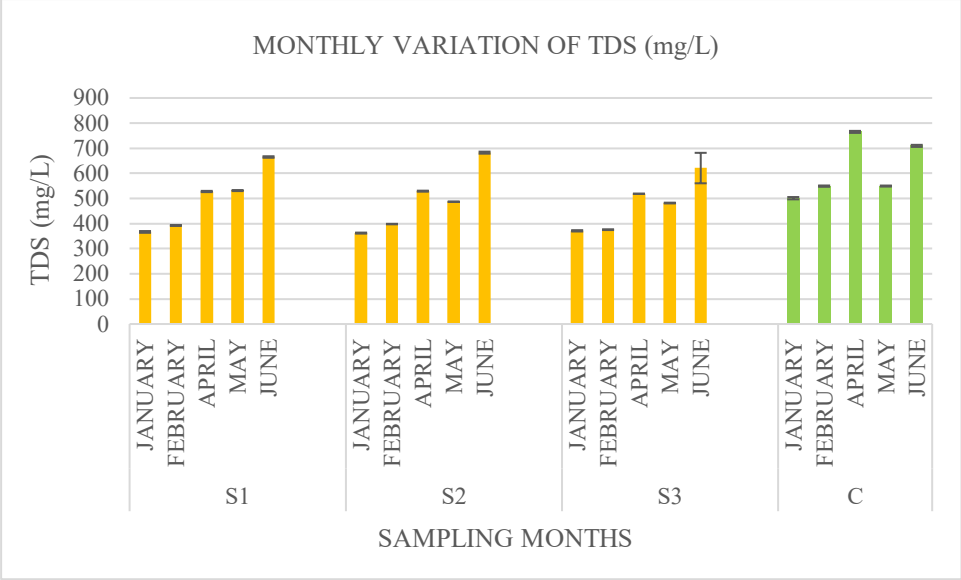


**FIGURE 8.3- Monthly Variation of TDS (mg/L) across Sampling Sites and Canal**

## Significance of TDS in the study-

TDS plays a vital role in regulating osmotic balance in aquatic organisms, particularly fish. Low TDS may lead to nutrient deficiency and poor productivity, while excessively high TDS can cause physiological stress and hinder fish growth. Hence, maintaining optimal TDS is essential for a balanced aquatic ecosystem. As per the BIS and CPCB guidelines, the acceptable TDS range for aquaculture lies between 300-800 mg/L.

## Monthly Variation of TDS-

Across all three aquaculture sites (S1, S2, S3), the TDS concentrations generally remained within a moderate range and showed a seasonal rise towards the summer months, likely due to increased evaporation and concentration of dissolved constituents. The Canal (C) however, consistently exhibited higher TDS levels, peaking in April and June. Elevated TDS values in the canal may be attributed to runoff from urban sources**,** sewage input, or reduced flushing and dilution. These high concentrations could affect the aquatic biota by influencing osmotic balance, especially in fish species sensitive to ionic changes in the water column. In this study, although TDS levels remained within the permissible range for aquaculture, the canal's upper values were approaching the higher limit, suggesting potential stress conditions, especially during summer months. Monitoring and management of ionic load and potential sources of salinity in the canal are crucial for sustainable fish farming and ecosystem health in the East Kolkata Wetlands.

## 8.4 Conductivity (µS)

This section evaluates the monthly and spatial variation of electrical conductivity across aquaculture sites (S1, S2, S3) and the adjoining canal (C) in the East Kolkata Wetlands. Conductivity reflects the concentration of dissolved ions in water and is directly correlated with Total Dissolved Solids (TDS). As the ionic strength of water increases, conductivity values rise, making it a crucial indicator for assessing water quality in fish farming systems.

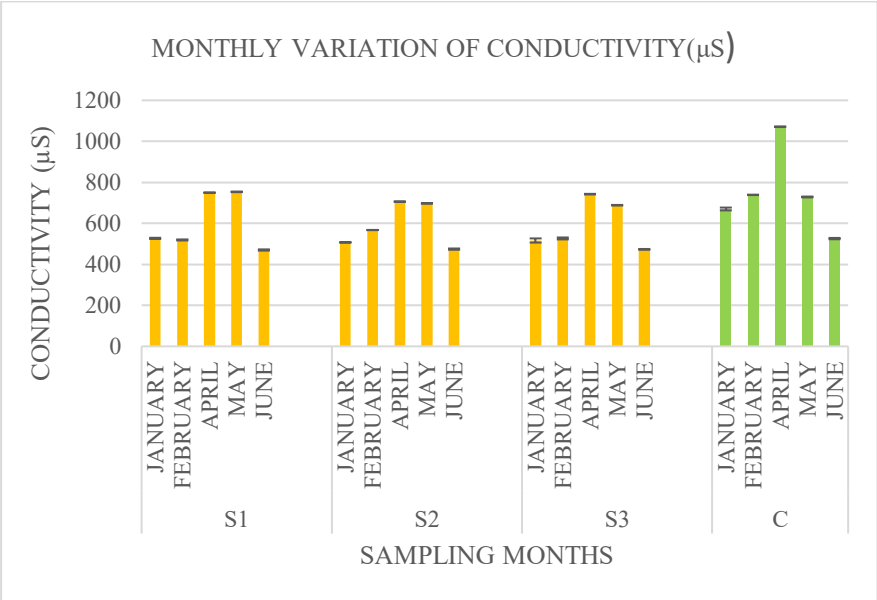


**FIGURE 8.4- Monthly Variation of Conductivity (µS) across Sampling Sites and Canal**

**Significance of Conductivity in the study-**

Conductivity plays an important role in regulating the osmotic balance of aquatic species, particularly fish. Elevated conductivity levels may result from the accumulation of dissolved salts and minerals due to factors such as evaporation, runoff, or sewage discharge. Excessive ionic concentration can lead to physiological stress in fish, hinder growth, affect reproductive capacity, and potentially reduce survival rates, especially in sensitive freshwater aquaculture systems. According to aquaculture water quality standards, the acceptable conductivity range for fish farming is typically between 100 to 1000 µS/cm. Exceeding this limit can create suboptimal conditions for aquatic life. Maintaining conductivity within this range ensures a balanced ionic environment that supports fish health and productivity.

**Monthly Variation of Conductivity-**

At Site S1, conductivity values gradually increased from January to a peak in May followed by a decline in June. This trend suggests the influence of summer evaporation and subsequent dilution by early monsoonal rainfall. Site S2 recorded values ranging from in January to a maximum range in April. Conductivity remained within permissible limits as per BIS and

CPCB guidelines, but showed a clear seasonal rise toward summer. Similarly, Site S3 exhibited an increase from January to April, before decreasing in June. This reflects a consistent seasonal conductivity pattern across the aquaculture sites. The Canal (C) showed the highest conductivity values, with a sharp peak in April and May. These values exceeded the upper acceptable limit for aquaculture in April, suggesting potential environmental stress for fish. This surge can be attributed to concentrated ionic loads during dry summer months, likely due to urban runoff, sewage discharge, and reduced water flow.

## 8.5 DO (mg/L)

This section examines the monthly and spatial variation in Dissolved Oxygen (DO) concentrations across three aquaculture sites (S1, S2, S3) and the adjacent canal (C) in the East Kolkata Wetlands. Dissolved Oxygen is a critical parameter in assessing water quality, as it directly influences the metabolic processes, survival, and growth of aquatic organisms, particularly fish. DO levels are indicative of the ecological health of a water body and are affected by factors such as temperature, organic load, and photosynthetic activity.

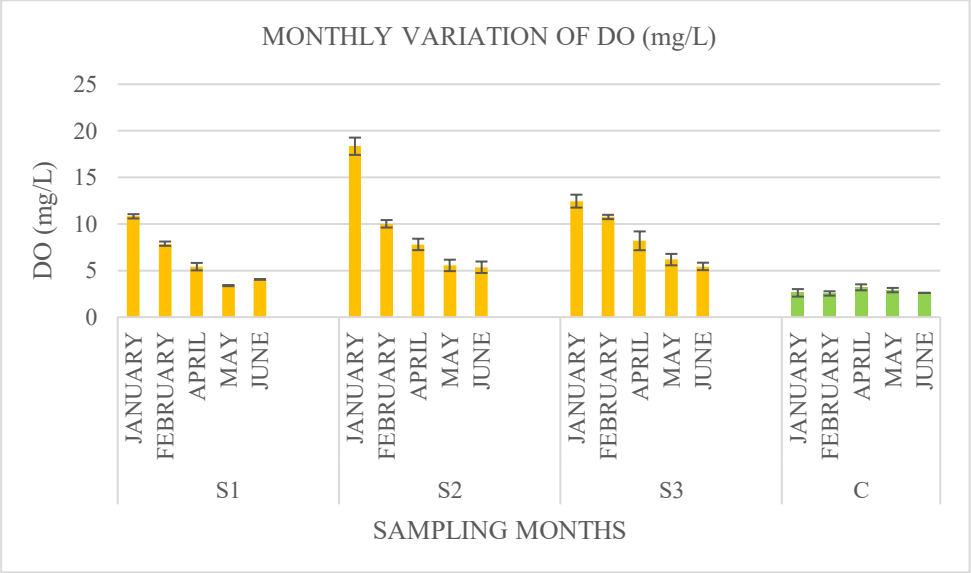


**FIGURE 8.5- Monthly Variation of DO (mg/L) across Sampling Sites and Canal**

**Significance of DO in the study-**

Dissolved Oxygen is indispensable for the survival of aquatic organisms. It plays a pivotal role in aerobic respiration and influences the decomposition of organic matter. In aquaculture systems, optimal DO levels are essential to support fish health, promote growth, and maintain an active microbial community that facilitates nutrient cycling. As per the Central Pollution Control Board (CPCB) and Bureau of Indian Standards (BIS), the minimum permissible limit of

DO for sustaining fish life is 5 mg/L. Concentrations below this threshold may lead to hypoxic stress, reduced feeding, and even fish mortality.

**Monthly Variation of DO-**

At all three aquaculture sites (S1, S2, S3), DO levels showed a clear decreasing trend from winter to summer. For instance, Site S1 recorded a high value of 10.81 mg/L in January, which declined progressively to a low of 3.38 mg/L in May, before slightly improving in June. A similar pattern was noted at Sites S2 and S3, where the highest values were seen in January (18.34 mg/L and 12.45 mg/L, respectively), decreasing steadily through May and June. These patterns can be attributed to elevated water temperatures in summer, which reduce the solubility of oxygen, along with increased microbial and algal respiration. The canal (C), however, exhibited consistently low DO levels across all months. January and June recorded values around 2.60 mg/L, with a marginal rise in April (3.2 mg/L). These figures are significantly below the permissible limit for aquaculture, indicating poor water quality and highlighting potential environmental stress in the canal zone. The degraded DO condition in the canal is likely due to high organic loading, limited aeration, and possibly sewage intrusion, especially during the warmer months.

## 8.6 BOD (mg/L)

This section evaluates the spatiotemporal variation of Biochemical Oxygen Demand (BOD) across three aquaculture sites (S1, S2, S3) and the adjacent canal (C) within the East Kolkata Wetlands. BOD is a critical water quality parameter that reflects the amount of oxygen required by microorganisms to decompose organic matter present in water. Elevated BOD levels indicate higher organic pollution, which can compromise aquatic life by depleting dissolved oxygen (DO) levels.

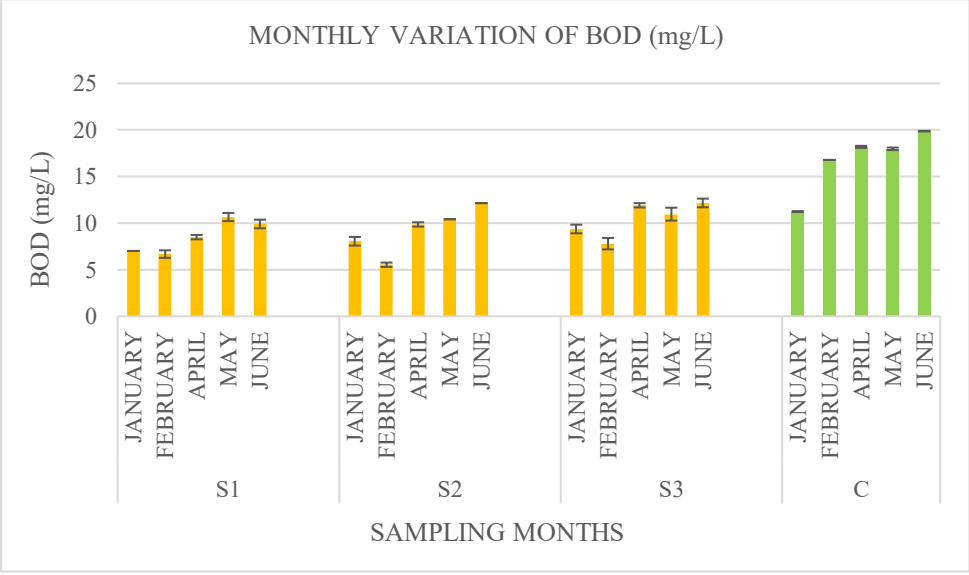


**FIGURE 8.6- Monthly Variation of BOD (mg/L) across sampling sites and canal**

**Significance of BOD in the study-**

BOD is inversely correlated with DO, serving as a key indicator of organic pollution and overall water quality. In aquaculture, elevated BOD can lead to oxygen depletion, resulting in stress or mortality in fish populations. As per the Central Pollution Control Board (CPCB) and Bureau of Indian Standards (BIS), the acceptable BOD concentration for fish culture should generally not exceed 3 mg/L. Values above this threshold can indicate organic overloading and pose a threat to fish health and pond productivity.

**Monthly Variation of BOD-**

Across the three aquaculture sites (S1, S2, and S3), BOD levels showed clear temporal trends, increasing progressively from winter to summer months. The lowest values were typically recorded in January and February, corresponding with cooler temperatures and reduced biological activity. In contrast, peak values appeared in May and June likely due to increased temperature-induced microbial activity and organic decomposition. At Site S1, BOD levels ranged from 6.67 mg/L in February to 10.65 mg/L in May. Site S2 exhibited a similar seasonal trend, with values peaking at 12.14 mg/L in June. Site S3 demonstrated the highest BOD value of 12.16 mg/L in June, indicating increasing organic load during warmer months. These elevated BOD levels during summer could adversely impact dissolved oxygen availability, stressing the aquatic fauna. The canal (C), however, exhibited significantly higher BOD values throughout the observation period, ranging from 11.24 mg/L in January to a maximum of 19.87 mg/L in June. The consistent and high BOD concentrations in the canal indicate severe organic pollution—likely from domestic sewage inflow, urban runoff, or unregulated waste discharge. These conditions render the canal water unsuitable for fish culture without adequate treatment or aeration support.

## 8.7 Free $CO_2$

This section presents the spatiotemporal variation in Free Carbon Dioxide (Free $CO_2$) concentrations across the three monitored aquaculture ponds (S1, S2, S3) and the adjoining canal (C) in the East Kolkata Wetlands. Free $CO_2$ in water arises from the respiration of aquatic organisms and the decomposition of organic matter, both of which release carbon dioxide into the system. It plays a critical role in aquatic chemistry by influencing pH, carbonate equilibrium, and acidification processes. Elevated levels of Free $CO_2$ are indicative of organic pollution and have direct implications for fish health and aquatic productivity.

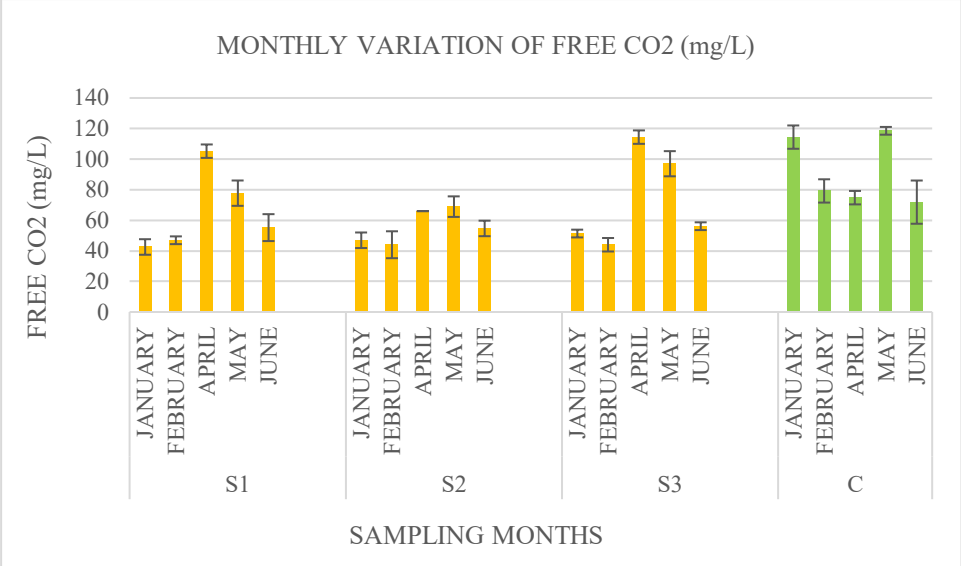


**FIGURE 8.7- Monthly Variation of Free $CO_2$ (mg/L) across sampling sites and canal**

## Significance of Free $CO_2$ in the study-

Free $CO_2$ is an essential water quality parameter that affects acid-base balance and buffering capacity in aquatic environments. High concentrations of dissolved carbon dioxide can lower pH by forming carbonic acid, thereby altering the chemical equilibrium of the system. This is particularly concerning for aquaculture, as both low pH and high $CO_2$ levels can impair fish respiration and reduce overall productivity. The presence of Free $CO_2$ in excess is often associated with increased organic matter and microbial activity, pointing toward pollution from decomposing biological waste or sewage inputs. As observed in the study, the temporal increase in Free $CO_2$ during summer months closely mirrors the pattern of Biological Oxygen Demand (BOD), further reinforcing the link between organic load and $CO_2$ buildup. According to aquaculture water quality guidelines, Free $CO_2$ levels exceeding 15 mg/L are considered stressful for most freshwater fish species. Persistent exposure to higher concentrations may result in respiratory distress, reduced oxygen uptake efficiency, and suppressed growth in cultured fish.

## Monthly Variation of Free $CO_2$-

S1 exhibited a sharp increase in Free $CO_2$ from 42.53 mg/L (January) to 105.2 mg/L (April), followed by elevated values in May (77.73 mg/L) and a slight decline in June (55.2 mg/L). This trend highlights the influence of summer temperature and organic decomposition. S2 followed a comparable seasonal pattern, with Free $CO_2$ rising from winter values around 44–46.93 mg/L to 68.93 mg/L in May, before slightly declining in June (54.67 mg/L). Notably, April recorded a dip (66 mg/L) despite the summer transition, likely due to local water dynamics. S3 demonstrated the highest site-level concentration in April (114.4 mg/L) and May (97 mg/L), underscoring intensified microbial respiration and poor buffering. The January and June values

remained significantly lower at 51.33 mg/L and 56.13 mg/L, respectively. The canal (C) consistently recorded the highest Free $CO_2$ values across all months, peaking at 118.53 mg/L in May, followed by 114.4 mg/L in January and 71.87 mg/L in June. The elevated and persistent levels indicate a greater degree of organic contamination, likely from urban runoff, sewage discharge, or stagnant water conditions.

## 8.8 Chloride

This section illustrates the monthly fluctuation of chloride ion concentration across the aquaculture ponds (S1, S2, S3) and the adjoining canal (C) in the EKW. Chloride, being a major anion present in water, plays an important role in maintaining osmotic and ionic balance in aquatic organisms. As a constituent of dissolved salts, chloride trends are often aligned with those of Total Dissolved Solids (TDS), particularly in systems experiencing evaporation, anthropogenic input, or salinity fluctuations.

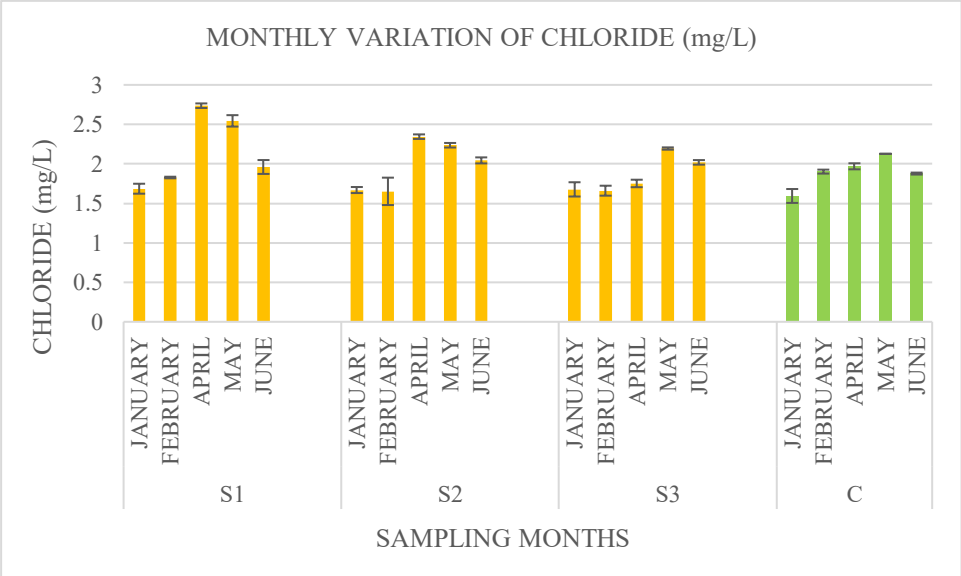


**FIGURE 8.8- Monthly Variation of Chloride (mg/L) across sampling sites and canal**

**Significance of Chloride in the study-**

Chloride is essential for maintaining the osmotic balance in aquatic organisms. However, excessive concentrations can disrupt the ionic regulation in fish, particularly in freshwater species, leading to physiological stress and lowered survival rates. According to aquaculture guidelines, the acceptable chloride concentration for freshwater fish ranges between 0.5 – 2.0 mg/L, with levels beyond this range posing risks for fish reproduction and health, particularly in closed aquaculture systems. In the present study, chloride patterns closely mirrored those of TDS, reinforcing the fact that both are reflective of salt accumulation and ionic concentration in

the water. This is particularly evident in the canal site (C), which consistently exhibited higher chloride values, possibly due to higher pollutant load, urban runoff, or stagnant flow conditions.

**Monthly Variation of Chloride-**

S1 displayed a progressive rise in chloride from January (1.69 mg/L) to a peak in April (2.74 mg/L), followed by a slight decline in June (1.96 mg/L). The peak corresponds to the pre-monsoon season, marked by high evaporation and reduced inflow, leading to salt concentration. S2 also recorded elevated levels during April (2.34 mg/L) and May (2.24 mg/L), aligning with the seasonal increase in ionic concentration due to thermal enrichment. S3, while generally consistent, showed a peak in May (2.19 mg/L), with comparatively lower values during the early months. Notably, this site maintained closer adherence to the acceptable threshold but exhibited moderate rises in the dry months. The Canal (C), in contrast to TDS and conductivity, showed relatively stable but gradually increasing values from January (1.59 mg/L) to May (2.13 mg/L), remaining slightly below the values recorded in the pond sites during peak months. This indicates lesser chloride accumulation in the canal compared to TDS—suggesting that other ions (like bicarbonates or nitrates) contribute more significantly to the canal's TDS profile than chloride alone.

## 8.9 Salinity

This section presents a comprehensive analysis of the monthly salinity levels across the aquaculture sites (S1, S2, S3) and the adjoining canal (C) in the East Kolkata Wetlands. Salinity, defined as the concentration of dissolved salts in water, is a fundamental parameter affecting aquatic life, especially fish physiology. Given that salinity is largely influenced by the presence of chloride ions, its pattern typically mirrors that of chloride distribution.

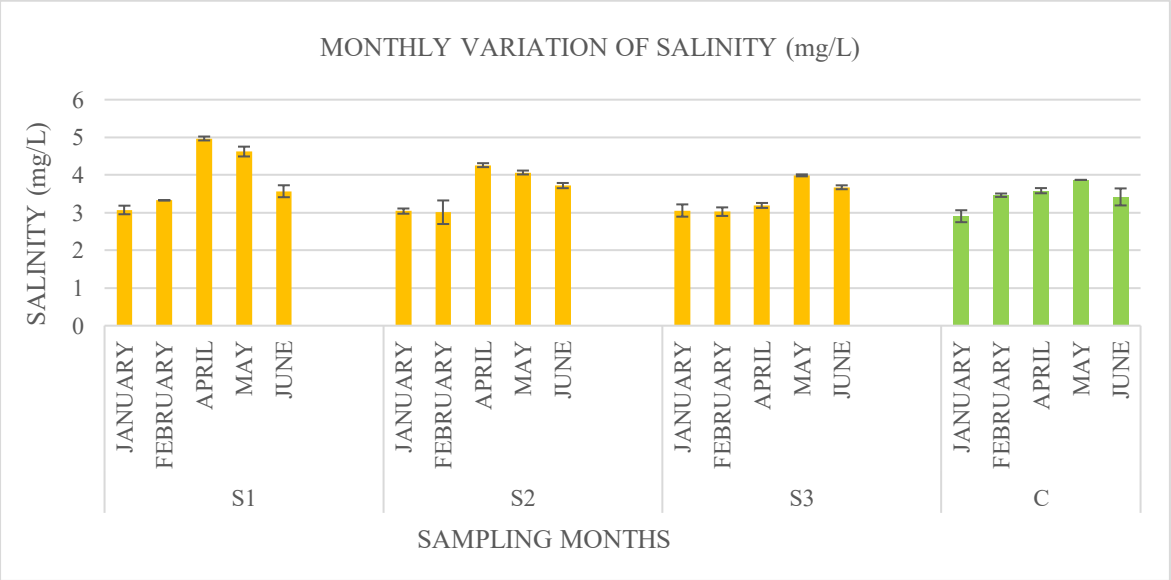


**FIGURE 8.9- Monthly Variation of Salinity (mg/L) across sampling sites and canal**

**Significance of Salinity in the study-**

Salinity significantly impacts osmoregulation in fish, which governs water and ion balance across membranes. Variations outside the optimal range can lead to stress, disrupted metabolism, and reduced growth. For freshwater aquaculture, a salinity range of 0.5–5 mg/L is generally considered acceptable, depending on species tolerance. Values approaching the upper threshold, especially during warmer months, may induce physiological stress in species less tolerant to saline fluctuations.

**Monthly Variation of Salinity-**

At Site S1, salinity levels ranged from 3.07 mg/L in January to a peak of 4.97 mg/L in April, reflecting a steady increase during the pre-monsoon dry season. The sharp rise during April and May can be attributed to high evaporation and reduced water inflow, which concentrate dissolved salts. A decrease in June (3.57 mg/L) corresponds with seasonal rainfall and dilution. Site S2 exhibited a similar pattern, starting from 3.04 mg/L in January and increasing to 4.26 mg/L in April and 4.06 mg/L in May, before tapering to 3.72 mg/L in June. This pattern reflects a strong seasonal influence and suggests similar hydrological conditions as S1. Site S3 demonstrated relatively moderate fluctuations, with salinity values remaining more stable across months. While an increase was observed from February (3.03 mg/L) to May (3.99 mg/L), the variation was less pronounced compared to S1 and S2, indicating more consistent water exchange or less evaporative stress. The Canal (C), interestingly, showed steadily increasing salinity from 2.91 mg/L in January to 3.86 mg/L in May, followed by a slight drop in June (3.42 mg/L). Despite being more exposed to runoff, the canal displayed salinity levels within acceptable limits, likely due to greater water volume and flow that buffers against ion concentration.

## 8.10 Alkalinity

This section presents the monthly fluctuation in alkalinity concentration across the aquaculture sites (S1, S2, S3) and the adjacent canal (C) in the East Kolkata Wetlands. Alkalinity, primarily governed by the presence of bicarbonates, carbonates, and occasionally hydroxides, serves as a buffering agent that stabilizes the pH of water. It plays a crucial role in supporting aquatic life by neutralizing acidic inputs and maintaining ecosystem balance.

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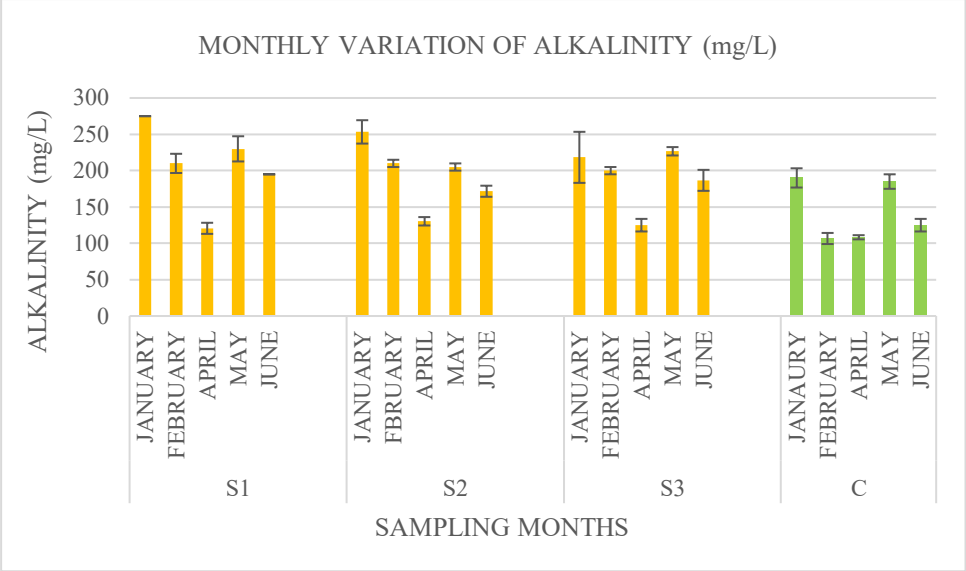


**FIGURE 8.10- Monthly Variation of Alkalinity (mg/L) across sampling sites and canal**

## Significance of Alkalinity in the study-

For aquaculture operations, an optimal alkalinity range of 75–200 mg/L is generally considered desirable for fish health, growth, and nutrient availability (Boyd, 1990). Alkalinity below 50 mg/L may lead to pH instability, while levels above 300 mg/L can impact the bioavailability of nutrients like phosphorus. In this study, most alkalinity values remained within or slightly above the optimal range, suggesting favourable buffering conditions for aquaculture. However, the sharp drop during April at all sites, especially at S1 (120.67 mg/L) and C (108.33 mg/L), calls for attention. These temporary declines could influence the uptake of essential nutrients and affect the physiological responses of farmed fish. Consistent monitoring of alkalinity is thus vital to maintain water quality, prevent pH swings, and support sustainable aquaculture practices throughout the seasonal cycle.

## Monthly Variation of Alkalinity-

The observed monthly variation in alkalinity aligns with seasonal hydrological changes in the wetland system. Higher values during January and May across sites can be attributed to reduced rainfall, minimal surface runoff, and increased mineral dissolution, which raises the concentration of bicarbonates and carbonates. In contrast, lower alkalinity levels in April and June suggest dilution effects due to pre-monsoon showers and enhanced surface flow. Among the sites, S1 consistently demonstrated higher alkalinity, suggesting either enhanced decomposition of organic matter**,** greater soil–water interactions, or local geochemical influences. In comparison, S2 and S3 exhibited more moderate levels, while the canal site showed the lowest alkalinity, likely due to higher water turnover and lesser interaction with sediment-bound carbonates**.** Alkalinity also correlates closely with pH and TDS stability. Its seasonal lows, particularly during April across all sites, highlight a potential period of reduced buffering capacity, which could make aquatic systems more vulnerable to pH fluctuations and associated stress on aquatic life.

## 8.11 Phosphate

This section presents the temporal distribution of phosphate concentrations across the three aquaculture sites (S1, S2, S3) and the adjacent canal (C) in the East Kolkata Wetlands. Phosphate is a critical nutrient in aquatic ecosystems but, in excess, serves as a key indicator of agricultural runoff and eutrophication risk particularly during pre-monsoon months when surface runoff intensifies due to preparatory agricultural practices.

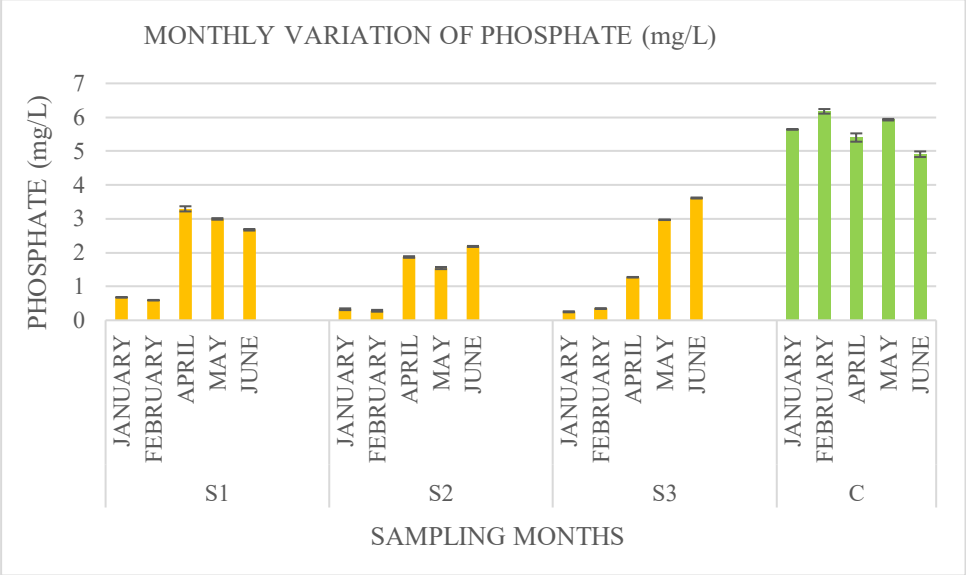


**FIGURE 8.11- Monthly Variation of Phosphate (mg/L) across sampling sites and canal**

### Significance of Phosphate in the study-

Phosphate, though essential for primary productivity, becomes detrimental when present in excess. The permissible limit for phosphate in aquaculture is typically <0.1 mg/L for freshwater systems, with values exceeding 0.3 mg/L considered eutrophic and potentially harmful due to algal blooms and oxygen depletion. In this context, the observed concentrations across all sites and especially the canal far exceed safe thresholds, signalling a potential ecological imbalance. Such high nutrient loads can reduce dissolved oxygen levels during decomposition, affecting fish health, survival, and overall productivity in culture systems.

### Monthly Variation of Phosphate-

Across all sampling stations, phosphate levels were markedly elevated during April to June, with the canal (C) consistently exhibiting the highest concentrations throughout the monitoring period. The peak value of 6.17 mg/L in February at the canal and sustained high values in subsequent months underscore the pronounced nutrient loading in these waters, likely attributable to the inflow of untreated runoff and domestic effluents. Site S1 showed a sharp increase in phosphate levels during the summer months, with concentrations peaking in April

(3.29 mg/L) and May (2.99 mg/L). This trend aligns with the onset of the agricultural season, where fertilizer use and dry weather conditions likely contribute to phosphate leaching into the aquaculture ponds. Similarly, Site S3 showed a gradual rise, culminating in the highest phosphate reading of 3.61 mg/L in June. Site S2 maintained comparatively lower concentrations but still showed a noticeable increase during summer, peaking at 2.18 mg/L in June. The spatial trend reveals a clear gradient, with the canal consistently exhibiting the highest phosphate values, followed by S1 and S3, while S2 generally reported the lowest levels. This gradient highlights the canal's role as a recipient of surface and agricultural runoff, intensifying nutrient loading especially in the warmer months.

## 8.12 TTC (Thermotolerant Coliform)

The monthly average TTC (Thermotolerant Coliform) concentrations across aquaculture sites S1, S2, S3, and the adjoining canal (C) are given below in form of graphical representation. A consistent trend of elevated TTC levels is observed during the summer months (April–June) across all sampling locations. The canal exhibits the highest TTC concentrations throughout the monitoring period, with values escalating from 600 CFU/mL in January to a peak of 1600 CFU/mL in June.

In contrast, the aquaculture sites (S1, S2, S3) record significantly lower TTC levels, ranging between 200 and 850 CFU/mL. For instance, in June, while the canal recorded 1600 CFU/mL, Site S2 displayed a much lower count of 800 CFU/mL, nearly half the value. This pattern suggests a substantial decrease—approximately tenfold—when water from the canal progresses towards the aquaculture zones.

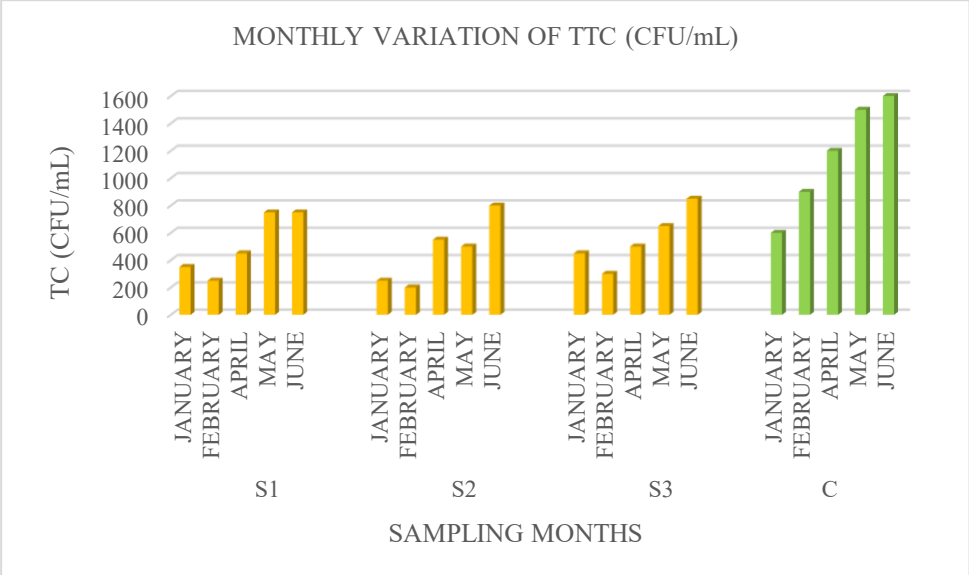


**FIGURE 8.12- Monthly Variation of TTC (CFU/mL) across sampling sites and canal**

**Significance of TTC in the study-**

The observed data illuminate potential health and ecological risks linked to microbiological pollution, especially in the canal stretch. Continuous monitoring and implementation of sanitation measures near inflow sources are essential to ensure microbial safety. Moreover, the substantial TTC attenuation from the canal to culture ponds suggests that natural purification plays a role, though not always sufficient during summer peaks. Effective wastewater management, especially during the pre-monsoon period, is vital to sustain water quality suitable for aquaculture in the East Kolkata Wetlands.

**Monthly Variation of TTC-**

Thermotolerant coliforms, incubated at 44°C, serve as crucial bioindicators of fecal contamination, primarily originating from warm-blooded animals. Their presence reflects the extent of animal or human waste intrusion into aquatic ecosystems. The conspicuously high TTC counts in canal waters imply a strong influence of untreated sewage or agricultural runoff, consistent with the canal's role as a conduit for drainage from surrounding urban and peri-urban catchments.

Seasonally, the pronounced rise in TTC levels during the summer months correlates with pre-monsoon environmental conditions. Elevated temperatures and reduced dilution due to minimal rainfall enhance microbial survival and concentration. This seasonal pattern further underscores the potential risk of microbial contamination during warmer periods.

The stark contrast in TTC levels between the canal and aquaculture sites supports the hypothesis of progressive microbial attenuation or dilution as water transitions from the canal to fish culture areas. Factors such as natural sedimentation, microbial competition, and water retention in pond systems likely contribute to this observed reduction.

According to the BIS and FAO guidelines, TTC counts for safe aquaculture practices should not exceed 1000 CFU/mL. In this study, while the canal frequently surpasses this threshold, aquaculture sites mostly remain within acceptable limits, suggesting moderate suitability for aquaculture in those areas. However, the values approaching the upper limit during peak summer months—particularly at S3 and S2 in June—warrant attention, as persistent exposure to such microbial loads can compromise fish health and market safety.

## 8.13 TC (Total Coliform)

This section outlines the monthly distribution of Total Coliform (TC) across aquaculture sites (S1, S2, S3) and the adjoining canal (C) in the East Kolkata Wetlands. TC is a widely used microbial indicator to assess the presence of faecal or sewage contamination in aquatic environments. It reflects the sanitary condition of the water and directly impacts its suitability for fish culture.

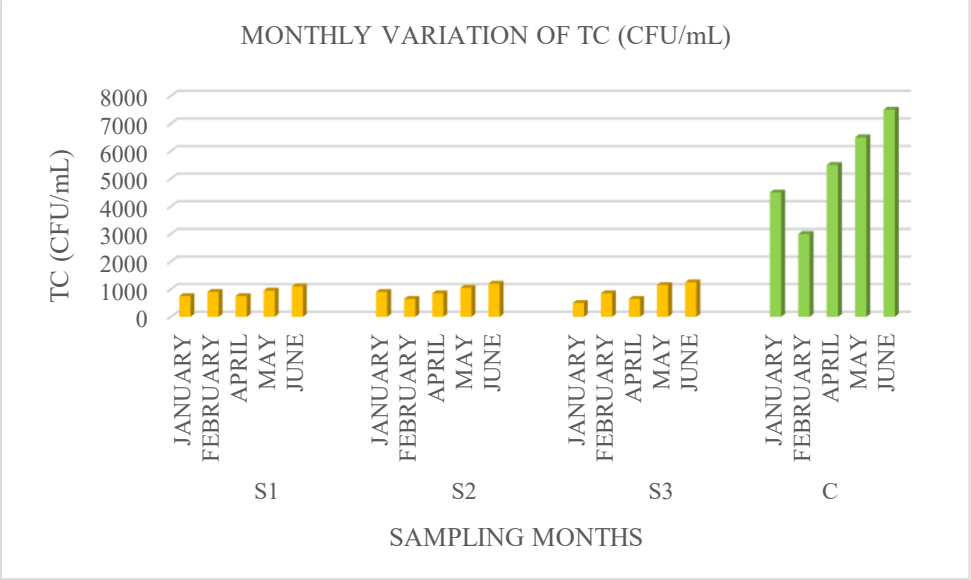


**FIGURE 8.13- Monthly Variation of TC (CFU/mL) across sampling sites and canal**

**Significance of TC in the study-**

The spatial reduction in TC from the canal to the aquaculture sites underscores the ecological importance of the East Kolkata Wetlands in naturally improving water quality. The data reinforces the need to regulate upstream sewage discharge and promote integrated wetland-aquaculture management practices to ensure sustainable fish farming and environmental safety.

**Monthly Variation of TC-**

TC levels at the canal (C) displayed notably elevated values throughout the monitoring period, ranging from 3000 CFU/mL in February to a peak of 7500 CFU/mL in June**.** In contrast, the aquaculture sites (S1, S2, S3) recorded comparatively lower and more consistent TC values. For instance, in S1, TC levels ranged between 750 CFU/mL in January and 1100 CFU/mL in June**.** S2 and S3 also exhibited a similar trend with values ranging between 650–1200 CFU/mL**,** with minor fluctuations observed across months. A general pattern of increasing TC concentration was observed during the summer months (April–June), likely linked to higher microbial activity and intensified runoff. The data also indicates a marked decline in TC levels nearly 6-10 fold as water flows from the canal to the aquaculture sites. The persistently high TC concentration in the canal highlights a significant level of sewage intrusion into the system. Such microbial loading may originate from untreated domestic waste, stormwater discharge, and surface runoff from densely populated areas. Total Coliform acts as an early warning parameter for potential health hazards in aquaculture operations, particularly in terms of fish disease outbreaks and contamination of harvested products.The observed reduction in TC concentration at the sites (S1–S3) as compared to the canal indicates a progressive improvement in water quality as the canal water is absorbed into the wetland environment. This suggests that the wetland filtration and assimilation processes play a crucial role in mitigating microbial loads. Although seasonal variability was marginal at the site level, a slight uptick

during the summer months could be attributed to the elevated temperature enhancing microbial proliferation and the increased load of organic matter from surface runoff. According to the BIS and FAO guidelines, water meant for aquaculture should ideally have TC levels below 1000 CFU/mL. The canal water exceeds this threshold by several folds, deeming it unsuitable for direct aquaculture use. However, water from the aquaculture sites, while occasionally exceeding this value, remains comparatively safer and may still be managed through periodic monitoring and appropriate water treatment.

## 8.14 Water Quality Index

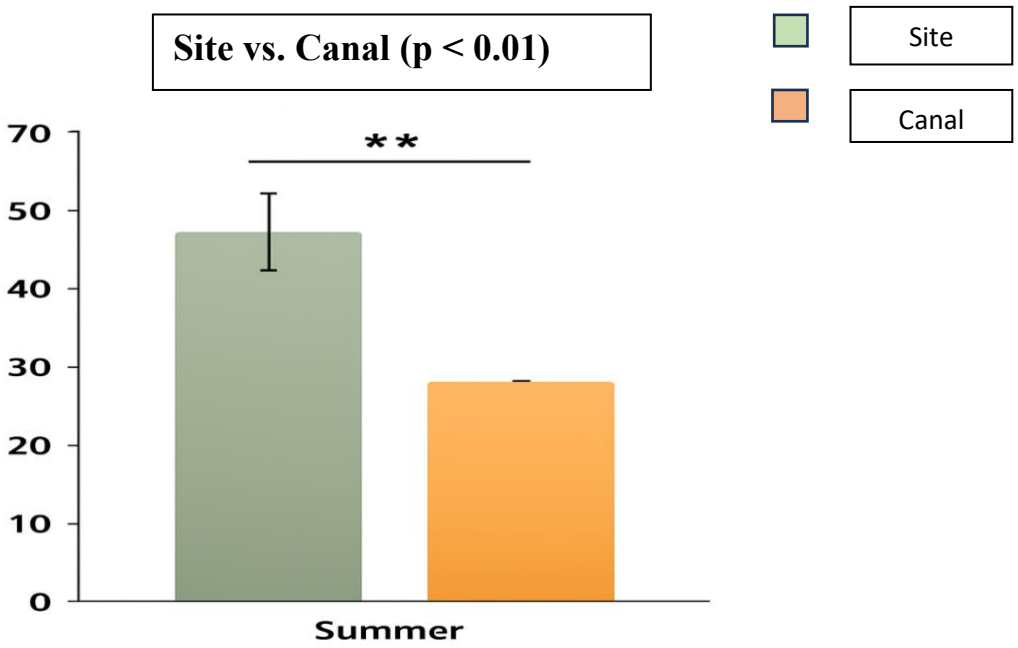


**FIGURE 8.14- Spatial Variation in Water Quality Index (WQI) for pond and canal waters during summer**

The Water Quality Index (WQI) provides an integrated measure of overall water quality by evaluating multiple physicochemical parameters. Based on the APHA 2024 guidelines, the WQI for each parameter was calculated using the formula:

**WQI parameter (p)**= (Measured value/Standard value)*100

**Overall WQI=** $\sum$WQI p/N,

where, N represents the number of water quality parameters assessed

According to the classification scheme, WQI values fall into four categories:

- **0–30**: Poor water quality
- **30–50**: Moderate
- **50–70**: Good
- **70+**: Excellent

A significant spatial variation in WQI values between pond and canal waters is seen during summer month (April to June) as depicted in Figure 8.14. The pond exhibited a considerably higher WQI (~50), indicating **good water quality**, while the canal showed a much lower WQI (~30), falling into the **moderate** to **poor** range. The error bars suggest greater variability in the pond data, possibly reflecting site-specific factors influencing water conditions. Statistical analysis revealed a highly significant difference (**$p < 0.01$**) between the two systems, highlighting the canal's reduced water quality during summer. This decline could be attributed to increased pollutant loading from runoff and limited water exchange, especially under high evaporation rates in the summer months. Overall, the WQI findings underscore that **pond systems are more suitable for aquaculture during summer** compared to canals. The marked variation also signals the need for **targeted water quality management in canal waters**, especially during warmer months.

# CHAPTER 09: CONCLUSION

This study offers a comprehensive assessment of the water quality in the aquaculture bheris of the East Kolkata Wetlands (EKW), with a specific focus on physico-chemical and microbiological parameters. The research was conducted over a six-month period, capturing seasonal and spatial variations across multiple sites. By analyzing parameters such as temperature, pH, dissolved oxygen (DO), biochemical oxygen demand (BOD), total dissolved solids (TDS), conductivity, free $CO_2$, and faecal coliform counts, the study aimed to evaluate the suitability of water for aquaculture and understand the underlying ecological dynamics of this Ramsar-recognized wetland ecosystem.

The findings indicate a notable improvement in the Water Quality Index (WQI) across several sites, which suggests enhanced ecological conditions and growing suitability for sustainable aquaculture. Particularly significant is the spatio-temporal variation observed in key water quality indicators. Seasonal factors such as temperature and rainfall, along with site-specific influences like sewage input, urban runoff, and agricultural discharge, led to dynamic fluctuations in parameters such as DO, BOD, phosphate, nitrate, and coliform levels. This variation underscores the need for localized and seasonal water quality management practices in the EKW.

One of the most encouraging outcomes of this study is the significant reduction in faecal coliform counts at multiple sites over the sampling period. This decline in microbial load reflects a possible natural recovery of the system, improved biological processing within the wetland, or

reduced pollutant inflow during specific seasons. The reduction of microbial contamination is critical for ensuring safe and healthy aquaculture practices, as excessive microbial presence poses risks to fish health and human consumers.

However, a key limitation of this study lies in the short sampling duration of six months, which restricted the ability to perform robust correlation and statistical analyses. As a result, deeper relationships between variables—such as DO and BOD, or microbial load and nutrient levels—could not be confidently established. This represents a methodological drawback that future research must address. A longer-term monitoring program, spanning at least 2 to 3 years, is necessary to enable seasonal modeling, time series analysis, and correlation-based studies. Such data will support more statistically sound conclusions and facilitate predictive modeling of water quality trends.

Looking ahead, this research lays the groundwork for several important future directions. With extended monitoring and sampling, it would be possible to develop predictive models that forecast water quality changes over a 5–6 year timeframe. These models could help anticipate pollution spikes, identify emerging threats to aquaculture, and inform better management strategies. Furthermore, microbial analysis can be expanded to isolate and identify specific pathogenic strains, which would not only enhance understanding of waterborne disease risks but also support the development of targeted interventions such as the use of probiotics, safe antibiotics, or biological controls.

The study also holds significant implications for policy and water resource management. By moving beyond conventional aquaculture practices and embracing innovative technologies, such as biofiltration, constructed wetlands, and microbial bioremediation, water quality can be further enhanced in an ecologically sustainable manner. The results of this research can guide policymakers and local stakeholders in implementing evidence-based interventions to safeguard the wetland's ecological functions while supporting its socioeconomic role in fish farming.

In conclusion, while the present research is limited in temporal scope, it provides valuable baseline data and critical insights into the evolving water quality of the East Kolkata Wetlands. It emphasizes the importance of integrated monitoring, microbial risk assessment, and adaptive aquaculture practices. With continued research and collaborative management, the EKW can serve as a model for sustainable wetland-aquaculture systems globally, balancing environmental conservation with community livelihoods.

# CHAPTER 10: APPENDIX (A)

## 10.1 Mean Temperature

| Site | Sample | Mean Temp (°C) | Standard Deviation |
|---|---|---|---|
| S1 | January | 21 | 0.1 |
| | February | 20.46666667 | 0.05774 |
| | April | 33.5 | 0.1 |
| | May | 33.7 | 0.2 |
| | June | 34.6333333 | 0.15275 |
| | | | |
| S2 | January | 20.1333333 | 0.11547 |
| | February | 19.7666667 | 0.05774 |
| | April | 34.0333333 | 0.05774 |
| | May | 33.9 | 0.1 |
| | June | 33.9666667 | 0.15275 |
| | | | |
| S3 | January | 20.9333333 | 0.05774 |
| | February | 21.1 | 0.1 |
| | April | 33.0333333 | 0.05774 |
| | May | 34.2 | 0.2 |
| | June | 32.0666667 | 0.20817 |
| | | | |
| C | January | 19.9 | 0.17321 |
| | February | 21.0333333 | 0.05774 |
| | April | 33.1 | 0.1 |
| | May | 31.0666667 | 0.11547 |
| | June | 30.9666667 | 0.05774 |

**TABLE 10.1- Monthly Variation of Mean Temp**

## 10.2 Mean pH

| Site | Sample | Mean pH | Standard deviation |
|---|---|---|---|
| S1 | January | 7.536667 | 0.005774 |
| | February | 7.823333 | 0.020817 |
| | April | 7.426667 | 0.020817 |
| | May | 8.29 | 0.01 |
| | June | 8.29 | 0.01 |
| | | | |
| S2 | January | 7.896667 | 0.015275 |
| | February | 7.71 | 0.036056 |
| | April | 7.76 | 0.02 |
| | May | 8.16 | 0.02 |
| | June | 8.16 | 0.02 |
| | | | |
| S3 | January | 7.81 | 0.01 |
| | February | 8.31 | 0.01 |
| | April | 8.086667 | 0.015275 |
| | May | 8.393333 | 0.030551 |
| | June | 8.393333 | 0.030551 |
| | | | |
| C | January | 6.39 | 0.01 |
| | February | 6.433333 | 0.025166 |
| | April | 6.283333 | 0.005774 |
| | May | 6.506667 | 0.050332 |
| | June | 6.506667 | 0.050332 |

**TABLE 10.2 Monthly Variation of Mean pH**

## 10.3 TDS

| Site | Sample | TDS (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 366.666667 | 3.51188 |
| | February | 392.333333 | 1.52753 |
| | April | 527.666667 | 2.51661 |
| | May | 531.333333 | 2.08167 |
| | June | 665 | 3 |
| | | | |
| S2 | January | 362 | 2 |
| | February | 398 | 1 |
| | April | 529 | 2 |
| | May | 487 | 1 |
| | June | 682.333333 | 4.04145 |
| | | | |
| S3 | January | 371 | 2.64575 |
| | February | 375.666667 | 1.52753 |
| | April | 518.666667 | 0.57735 |
| | May | 481.666667 | 1.52753 |
| | June | 621 | 60.6548 |
| | | | |
| C | January | 501 | 4.58258 |
| | February | 549 | 2.64575 |
| | April | 765 | 4 |
| | May | 549.333333 | 2.08167 |
| | June | 709.333333 | 4.16333 |

**TABLE 10.3- Monthly Variation of TDS**

## 10.4 Conductivity

| Site | Sample | Conductivity (µS) | Standard deviation |
|---|---|---|---|
| S1 | January | 526.6666667 | 2.88675 |
| | February | 519 | 2.64575 |
| | April | 749.6666667 | 1.52753 |
| | May | 753.6666667 | 1.52753 |
| | June | 470 | 3 |
| | | | |
| S2 | January | 507 | 2 |
| | February | 567.3333333 | 0.57735 |
| | April | 706 | 1 |
| | May | 697.3333333 | 1.1547 |
| | June | 474.3333333 | 3.05505 |
| S3 | January | 516.3333333 | 10.2632 |
| | February | 526.3333333 | 4.72582 |
| | April | 742.3333333 | 1.52753 |
| | May | 688.3333333 | 1.52753 |
| | June | 473 | 2 |
| | | | |
| C | January | 670 | 7 |
| | February | 739 | 1 |
| | April | 1071.333333 | 1.52753 |
| | May | 728.6666667 | 2.08167 |
| | June | 525.6666667 | 3.21455 |

**TABLE-10.4 – Monthly Variation of Conductivity**

## 10.5 DO

| Site | Sample | DO (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 10.819277 | 0.231867578 |
| | February | 7.8821954 | 0.231867578 |
| | April | 5.4176707 | 0.401606426 |
| | May | 3.3868809 | 0.061346395 |
| | June | 4.0481928 | 0.028729027 |
| | | | |
| S2 | January | 18.340027 | 0.927470312 |
| | February | 10.016064 | 0.401606426 |
| | April | 7.8045515 | 0.613463948 |
| | May | 5.5528782 | 0.613463948 |
| | June | 5.3547523 | 0.613463948 |
| | | | |
| S3 | January | 12.449799 | 0.695602734 |
| | February | 10.749665 | 0.231867578 |
| | April | 8.1900937 | 1.010687341 |
| | May | 6.1753681 | 0.613463948 |
| | June | 5.4417671 | 0.401606426 |
| | | | |
| C | January | 2.6064257 | 0.401606426 |
| | February | 2.5435074 | 0.231867578 |
| | April | 3.2 | 0.3211 |
| | May | 2.9 | 0.235666 |
| | June | 2.6064257 | 8.07644E-15 |

**TABLE 10.5- Monthly Variation of DO**

## 10.6 BOD

| Site | Sample | BOD (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 7.0160643 | 0 |
| | February | 6.6773762 | 0.40160643 |
| | April | 8.49563 | 0.23421 |
| | May | 10.654232 | 0.43892 |
| | June | 9.9062918 | 0.46373516 |
| | | | |
| S2 | January | 8.0562249 | 0.46373516 |
| | February | 5.5354752 | 0.23186758 |
| | April | 9.8594378 | 0.23186758 |
| | May | 10.423173 | 0 |
| | June | 12.149933 | 1.0296E-15 |
| | | | |
| S3 | January | 9.3708166 | 0.46373516 |
| | February | 7.7965194 | 0.61346395 |
| | April | 11.914324 | 0.23186758 |
| | May | 10.962517 | 0.69560273 |
| | June | 12.165997 | 0.46373516 |
| | | | |
| C | January | 11.243476 | 0.00423165 |
| | February | 16.784354 | 0.00324164 |
| | April | 18.187435 | 0.10884536 |
| | May | 17.976352 | 0.135658 |
| | June | 19.874524 | 0.03241566 |

**TABLE 10.6- Monthly Variation of BOD**

## 10.7 Free CO2

| Site | Sample | Free CO2 (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 42.53333333 | 5.08068 |
| | February | 46.93333333 | 2.54034 |
| | April | 105.2 | 4.4 |
| | May | 77.73333333 | 8.28804 |
| | June | 55.2 | 8.8 |
| | | | |
| S2 | January | 46.93333333 | 5.08068 |
| | February | 44 | 8.8 |
| | April | 66 | 0 |
| | May | 68.93333333 | 6.72111 |
| | June | 54.66666667 | 5.08068 |
| | | | |
| S3 | January | 51.33333333 | 2.54034 |
| | February | 44 | 4.4 |
| | April | 114.4 | 4.4 |
| | May | 97 | 8.23 |
| | June | 56.13333333 | 2.54034 |
| | | | |
| C | January | 114.4 | 7.62102 |
| | February | 79.2 | 7.62102 |
| | April | 74.8 | 4.4 |
| | May | 118.5333333 | 2.54034 |

**TABLE 10.7- Monthly Variation of Free CO2**

## 10.8 Chloride

| Site | Sample | Chloride (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 1.685185 | 0.06298449 |
| | February | 1.8270075 | 0.01 |
| | April | 2.73634 | 0.02889927 |
| | May | 2.5444625 | 0.07224817 |
| | June | 1.9604875 | 0.08789369 |
| | | | |
| S2 | January | 1.6685 | 0.03823014 |
| | February | 1.651815 | 0.17339561 |
| | April | 2.3442425 | 0.02889927 |
| | May | 2.23579 | 0.02889927 |
| | June | 2.0439125 | 0.03823014 |
| | | | |
| S3 | January | 1.6768425 | 0.09023793 |
| | February | 1.6601575 | 0.06298449 |
| | April | 1.751925 | 0.04740751 |
| | May | 2.1940775 | 0.01444963 |
| | June | 2.018885 | 0.02889927 |
| | | | |
| C | January | 1.5934175 | 0.08789369 |
| | February | 1.90209 | 0.0250275 |
| | April | 1.96883 | 0.03823014 |
| | May | 2.1273375 | 0 |
| | June | 1.8770625 | 0.01251375 |

**TABLE 10.8- Monthly Variation of Chloride**

## 10.9 Salinity

| Site | Sample | Salinity (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | **3.071758925** | **0.113687** |
| | February | 3.327748538 | 3.14E-16 |
| | April | 4.9690937 | 0.052163 |
| | May | 4.622754813 | 0.130408 |
| | June | 3.568679938 | 0.158648 |
| | | | |
| S2 | January | 3.0416425 | 0.069005 |
| | February | 3.011526075 | 0.312979 |
| | April | 4.261357713 | 0.052163 |
| | May | 4.06560095 | 0.052163 |
| | June | 3.719262063 | 0.069005 |
| | | | |
| S3 | January | 3.056700713 | 0.162879 |
| | February | 3.026584288 | 0.113687 |
| | April | 3.192224625 | 0.066071 |
| | May | 3.990309888 | 0.026082 |
| | June | 3.674087425 | 0.052163 |
| | | | |
| C | January | 2.906118588 | 0.158648 |
| | February | 3.46327245 | 0.045175 |
| | April | 3.58373815 | 0.069005 |
| | May | 3.869844188 | 0 |
| | June | 3.418097813 | 0.225873 |

**TABLE 10.9- Monthly Variation of Salinity**

## 10.10 Alkalinity

| Site | Sample | Alkalinity (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 275 | 0 |
| | February | 210 | 13.22875656 |
| | April | 120.6666667 | 7.637626158 |
| | May | 230 | 17.32050808 |
| | June | 195 | 3.48093E-14 |
| | | | |
| S2 | January | 253.3333333 | 16.07275127 |
| | February | 210 | 5 |
| | April | 130.3333333 | 5.773502692 |
| | May | 205 | 5 |
| | June | 171.6666667 | 7.637626158 |
| | | | |
| S3 | January | 218.3333333 | 35.11884584 |
| | February | 200 | 5 |
| | April | 125 | 8.660254038 |
| | May | 226.6666667 | 5.773502692 |
| | June | 186.6666667 | 14.43375673 |
| | | | |
| C | January | 190 | 13.22875656 |
| | February | 106.6666667 | 7.637626158 |
| | April | 108.3333333 | 2.886751346 |
| | May | 185 | 10 |
| | June | 125 | 8.660254038 |

**TABLE 10.10- Monthly Variation of Alkalinity**

## 10.11 Phosphate

| Site | Sample | Phosphate (mg/L) | Standard deviation |
|---|---|---|---|
| S1 | January | 0.679235797 | 0.00871 |
| | February | 0.593765711 | 0.00871 |
| | April | 3.293614882 | 0.07592 |
| | May | 2.995977878 | 0.02304 |
| | June | 2.675213675 | 0.02304 |
| | | | |
| S2 | January | 0.327300151 | 0.02612 |
| | February | 0.282051282 | 0.02612 |
| | April | 1.870789341 | 0.02304 |
| | May | 1.543991956 | 0.0314 |
| | June | 2.182503771 | 0.01508 |
| | | | |
| S3 | January | 0.25188537 | 0.01508 |
| | February | 0.347410759 | 0.00871 |
| | April | 1.272498743 | 0.00871 |
| | May | 2.971845148 | 0.00871 |
| | June | 3.61387632 | 0.01508 |
| | | | |
| C | January | 5.641025641 | 0 |
| | February | 6.174459527 | 0.06801 |
| | April | 5.400201106 | 0.12284 |
| | May | 5.928104575 | 0.02304 |
| | June | 4.907491202 | 0.08307 |

**TABLE 10.11- Monthly Variation of Phosphate**

## 10.12 TTC

| Site | Sample | TTC (CFU/mL) |
|---|---|---|
| S1 | January | 350 |
| | February | 250 |
| | April | 450 |
| | May | 750 |
| | June | 750 |
| | | |
| S2 | January | 250 |
| | February | 200 |
| | April | 550 |
| | May | 500 |
| | June | 800 |
| | | |
| S3 | January | 450 |
| | February | 300 |
| | April | 500 |
| | May | 650 |
| | June | 850 |
| | | |
| C | January | 600 |
| | February | 900 |
| | April | 1200 |
| | May | 1500 |
| | June | 1600 |

**TABLE 10.12 – Monthly Variation of TTC**

# 10.13 TC

| Site | Sample | TC (CFU/mL) |
|---|---|---|
| S1 | January | 750 |
| | February | 900 |
| | April | 750 |
| | May | 950 |
| | June | 1100 |
| | | |
| S2 | January | 900 |
| | February | 650 |
| | April | 850 |
| | May | 1050 |
| | June | 1200 |
| | | |
| S3 | January | 500 |
| | February | 850 |
| | April | 650 |
| | May | 1150 |
| | June | 1250 |
| | | |
| C | January | 4500 |
| | February | 3000 |
| | April | 5500 |
| | May | 6500 |
| | June | 7500 |

**TABLE 10.13- Monthly Variation of TC**

# APPENDIX (B)

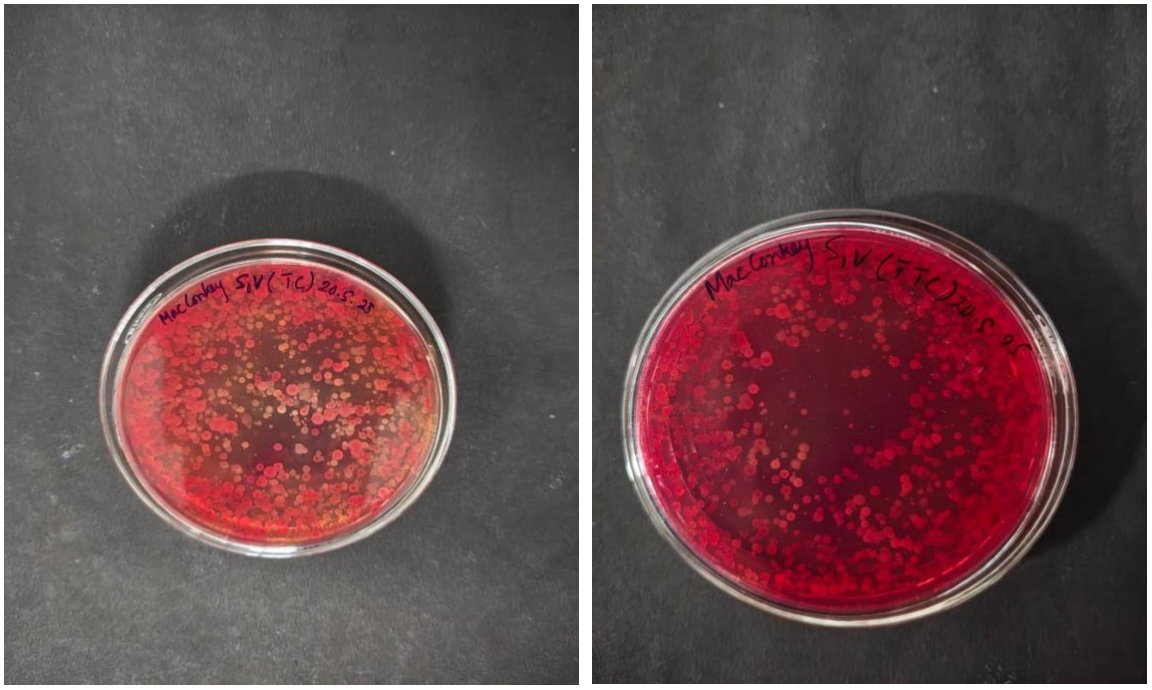

**FIGURE 10.1 (a) MacConkey Agar-Site 1 (TC) (b) MacConkey Agar-Site 1 (TTC)**

**FIGURE 10.2 (a) MacConkey Agar-Site 2 (TC) (b) MacConkey Agar-Site 2 (TTC)**

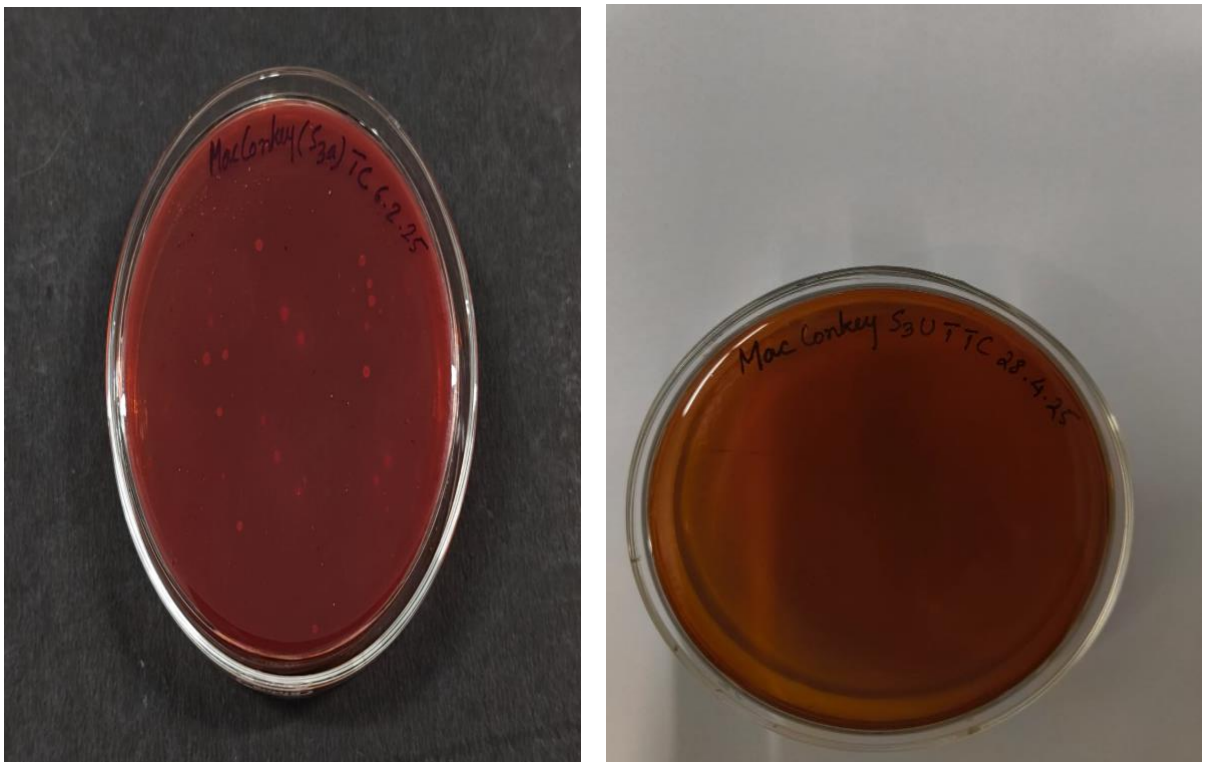


**FIGURE 10.3 (a) MacConkey Agar-Site 3 (TC) (b) MacConkey Agar-Site 2 (TTC)**

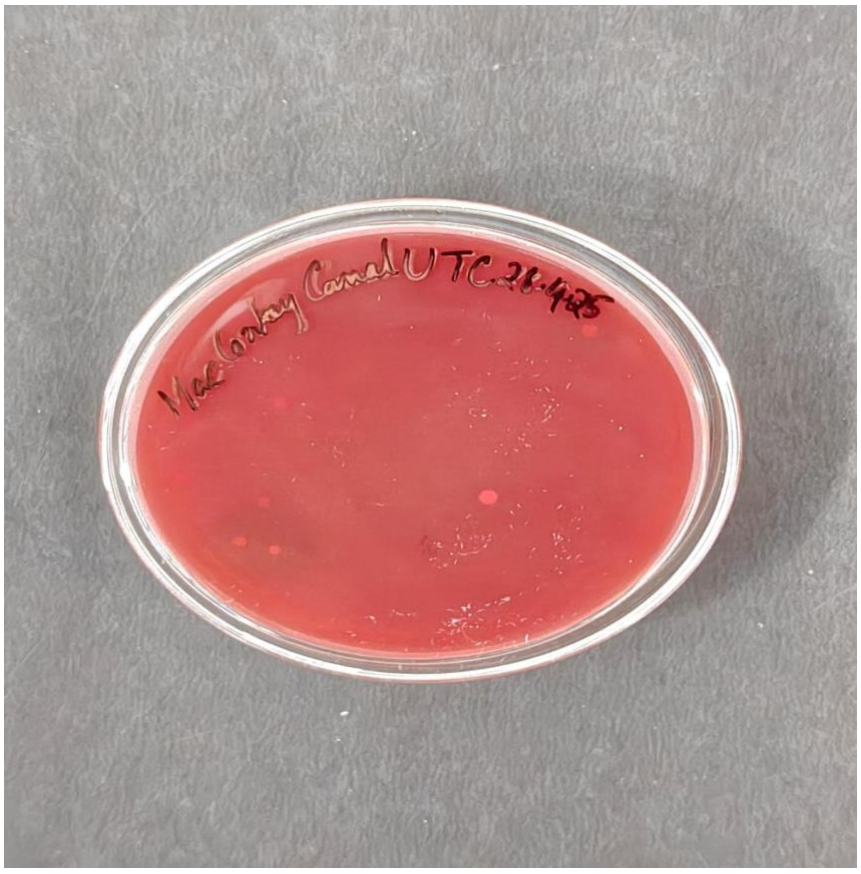


**FIGURE 10.4 (a) MacConkey Agar Canal (TC)**

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